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(54) Title: HETEROCYCLIC INHIBITORS OF P38			
(57) Abstract <p>The present invention relates to inhibitors of p38, a mammalian protein kinase involved in cell proliferation, cell death and response to extracellular stimuli. The invention also relates to methods for producing these inhibitors. The invention also provides pharmaceutical compositions comprising the inhibitors of the invention and methods of utilizing those compositions in the treatment and prevention of various disorders.</p>			

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HETEROCYCLIC INHIBITORS OF P38

TECHNICAL FIELD OF INVENTION

The present invention relates to inhibitors of p38, a mammalian protein kinase involved in cell proliferation, cell death and response to extracellular stimuli. The invention also relates to methods for producing these inhibitors. The invention also provides pharmaceutical compositions comprising the inhibitors of the invention and methods of utilizing those compositions in the treatment and prevention of various disorders.

BACKGROUND OF THE INVENTION

Protein kinases are involved in various cellular responses to extracellular signals. Recently, a family of mitogen-activated protein kinases (MAPK) has been discovered. Members of this family are Ser/Thr kinases that activate their substrates by phosphorylation [B. Stein et al., Ann. Rep. Med. Chem., 31, pp. 289-98 (1996)]. MAPKs are themselves activated by a variety of signals including growth factors, cytokines, UV radiation, and stress-inducing agents.

One particularly interesting MAPK is p38. p38, also known as cytokine suppressive anti-inflammatory drug binding protein (CSBP) and RK, was isolated from murine pre-B cells that were transfected with the lipopolysaccharide (LPS) receptor, CD14, and induced with LPS. p38 has since been isolated and sequenced, as has the cDNA encoding it in humans and mouse. Activation of p38 has been observed in cells stimulated by stress, such as treatment of lipopolysaccharides (LPS), UV,

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anisomycin, or osmotic shock, and by cytokines, such as IL-1 and TNF.

Inhibition of p38 kinase leads to a blockade on the production of both IL-1 and TNF. IL-1 and TNF stimulate the production of other proinflammatory cytokines such as IL-6 and IL-8 and have been implicated in acute and chronic inflammatory diseases and in post-menopausal osteoporosis [R. B. Kimble et al., Endocrinol., 136, pp. 3054-61 (1995)].

Based upon this finding, it is believed that p38, along with other MAPKs, have a role in mediating cellular response to inflammatory stimuli, such as leukocyte accumulation, macrophage/monocyte activation, tissue resorption, fever, acute phase responses and neutrophilia. In addition, MAPKs, such as p38, have been implicated in cancer, thrombin-induced platelet aggregation, immunodeficiency disorders, autoimmune diseases, cell death, allergies, osteoporosis and neurodegenerative disorders. Inhibitors of p38 have also been implicated in the area of pain management through inhibition of prostaglandin endoperoxide synthase-2 induction. Other diseases associated with IL-1, IL-6, IL-8 or TNF overproduction are set forth in WO 96/21654.

Others have already begun trying to develop drugs that specifically inhibit MAPKs. For example, PCT publication WO 95/31451 describes pyrazole compounds that inhibit MAPKs, and, in particular, p38. However, the efficacy of these inhibitors *in vivo* is still being investigated.

Accordingly, there is still a great need to develop other potent inhibitors of p38, including p38-

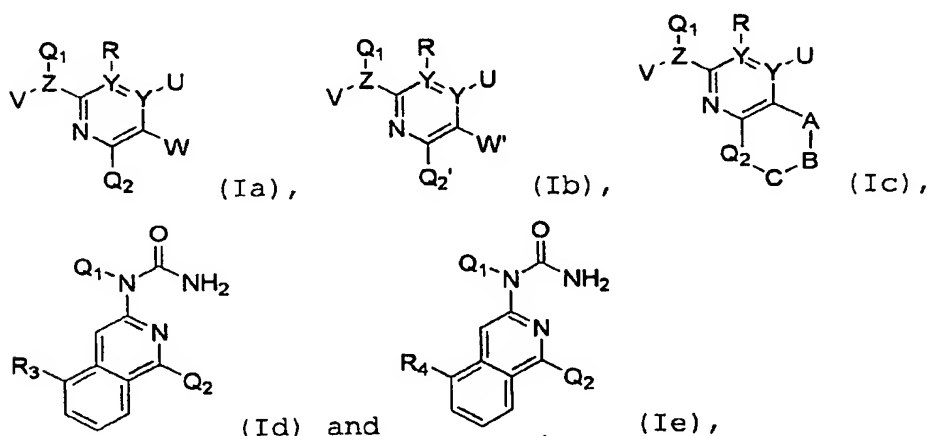
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specific inhibitors, that are useful in treating various conditions associated with p38 activation.

SUMMARY OF THE INVENTION

The present invention addresses this problem by providing compounds that demonstrate strong inhibition of p38.

These compounds have the general formula:



wherein each of Q₁ and Q₂ are independently selected from a phenyl or 5-6 membered aromatic heterocyclic ring system, or a 8-10 membered bicyclic ring system comprising aromatic carbocyclic rings, aromatic heterocyclic rings or a combination of an aromatic carbocyclic ring and an aromatic heterocyclic ring.

A heterocyclic ring system or a heterocyclic ring contains 1 to 4 heteroatoms, which are independently selected from N, O, S, SO and SO₂.

The rings that make up Q₁ are substituted with 1 to 4 substituents, each of which is independently selected from halo; C₁-C₃ alkyl optionally substituted with NR'₂, OR', CO₂R' or CONR'₂; O-(C₁-C₃)-alkyl

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optionally substituted with NR'_2 , OR' , $\text{CO}_2\text{R}'$ or CONR'_2 ;
 NR'_2 ; OCF_3 ; CF_3 ; NO_2 ; $\text{CO}_2\text{R}'$; CONR' ; SR' ; $\text{S}(\text{O}_2)\text{N}(\text{R}')_2$; SCF_3 ;
 CN ; $\text{N}(\text{R}')\text{C}(\text{O})\text{R}^4$; $\text{N}(\text{R}')\text{C}(\text{O})\text{OR}^4$; $\text{N}(\text{R}')\text{C}(\text{O})\text{C}(\text{O})\text{R}^4$;
 $\text{N}(\text{R}')\text{S}(\text{O}_2)\text{R}^4$; $\text{N}(\text{R}')\text{R}^4$; $\text{N}(\text{R}^4)_2$; OR^4 ; $\text{OC}(\text{O})\text{R}^4$; $\text{OP}(\text{O})_3\text{H}_2$; or
 5 $\text{N}=\text{C}-\text{N}(\text{R}')_2$.

The rings that make up Q_2 are optionally substituted with up to 4 substituents, each of which is independently selected from halo; C_1 - C_3 straight or branched alkyl optionally substituted with NR'_2 , OR' ,
 10 $\text{CO}_2\text{R}'$, $\text{S}(\text{O}_2)\text{N}(\text{R}')_2$, $\text{N}=\text{C}-\text{N}(\text{R}')_2$, R^3 , or CONR'_2 ; $\text{O}-(\text{C}_1-\text{C}_3)$ -alkyl; $\text{O}-(\text{C}_1-\text{C}_3)$ -alkyl optionally substituted with NR'_2 , OR' , $\text{CO}_2\text{R}'$, $\text{S}(\text{O}_2)\text{N}(\text{R}')_2$, $\text{N}=\text{C}-\text{N}(\text{R}')_2$, R^3 , or CONR'_2 ; NR'_2 ; OCF_3 ; CF_3 ; NO_2 ; $\text{CO}_2\text{R}'$; CONR' ; R^3 ; OR^3 ; NR^3 ; SR^3 ; $\text{C}(\text{O})\text{R}^3$; $\text{C}(\text{O})\text{N}(\text{R}')\text{R}^3$; $\text{C}(\text{O})\text{OR}^3$; SR' ; $\text{S}(\text{O}_2)\text{N}(\text{R}')_2$; SCF_3 ; $\text{N}=\text{C}-\text{N}(\text{R}')_2$;
 15 or CN .

Q_2' is selected from phenyl or a 5-6 member aromatic heterocyclic ring optionally substituted with 1-3 substituents, each of which is independently selected from halogen; C_1 - C_3 alkyl optionally substituted with
 20 NR'_2 , OR' , $\text{CO}_2\text{R}'$, CONR'_2 , or $\text{O}-\text{P}(\text{O}_3)\text{H}_2$; $\text{O}-(\text{C}_2-\text{C}_3)$ -alkyl optionally substituted with NR'_2 , OR' , $\text{CO}_2\text{R}'$, CONR'_2 , or $\text{OP}(\text{O}_3)\text{H}_2$; OCF_3 ; CF_3 ; OR^4 ; $\text{O}-\text{CO}_2\text{R}^4$; $\text{O}-\text{P}(\text{O}_3)\text{H}_2$; $\text{CO}_2\text{R}'$; CONR' ; SR' ; $\text{S}(\text{O}_2)\text{N}(\text{R}')_2$; SCF_3 ; CN ; $\text{N}(\text{R}')\text{C}(\text{O})\text{R}^4$; $\text{N}(\text{R}')\text{C}(\text{O})\text{OR}^4$; $\text{N}(\text{R}')\text{C}(\text{O})\text{C}(\text{O})\text{R}^4$; $\text{N}(\text{R}')\text{S}(\text{O}_2)\text{R}^4$; $\text{N}(\text{R}')\text{R}^4$; $\text{N}(\text{R}^4)_2$; OR^4 ;
 25 $\text{OC}(\text{O})\text{R}^4$; $\text{OP}(\text{O})_3\text{H}_2$; or $\text{N}=\text{C}-\text{N}(\text{R}')_2$; provided that Q_2' is not phenyl optionally substituted 1 to 3 substituents independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl.

R' is selected from hydrogen; (C_1-C_3) -alkyl;
 30 (C_2-C_3) -alkenyl or alkynyl; phenyl or phenyl substituted

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with 1 to 3 substituents independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl; or a 5-6 membered heterocyclic ring system optionally substituted with 1 to 3 substituents
 5 independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl.

R^3 is selected from 5-8 membered aromatic or non-aromatic carbocyclic or heterocyclic ring systems each optionally substituted with R' , R^4 , $-C(O)R'$, $-C(O)R^4$,
 10 $-C(O)OR^4$ or $-J$; or an 8-10 membered bicyclic ring system comprising aromatic carbocyclic rings, aromatic heterocyclic rings or a combination of an aromatic carbocyclic ring and an aromatic heterocyclic ring each optionally substituted with R' , R^4 , $-C(O)R'$, $-C(O)R^4$, $-$
 15 $C(O)OR^4$ or $-J$.

R^4 is (C_1-C_4) -straight or branched alkyl optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, or $SO_2N(R^2)_2$; or a 5-6 membered carbocyclic or heterocyclic ring system optionally substituted with
 20 $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, or $SO_2N(R^2)_2$.

R^5 is selected from hydrogen; (C_1-C_3) -alkyl optionally substituted with R^3 ; (C_2-C_3) -alkenyl or alkynyl each optionally substituted with R^3 ; phenyl or phenyl substituted with 1 to 3 substituents independently
 25 selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl; or a 5-6 membered heterocyclic ring system optionally substituted with 1 to 3 substituents independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl.

30 W is selected from $N(R^2)SO_2-N(R^2)_2$; $N(R^2)SO_2-N(R^2)(R^3)$; $N(R^2)C(O)-OR^2$; $N(R^2)C(O)-N(R^2)_2$; $N(R^2)C(O)-$

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$N(R^2)(R^3)$; $N(R^2)C(O)-R^2$; $N(R^2)_2$; $C(O)-R^2$; $CH(OH)-R^2$; $C(O)-N(R^2)_2$; $C(O)-OR^2$; J; or (C_1-C_4) straight or branched alkyl optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, R^3 , $SO_2N(R^2)_2$, $OC(O)R^2$, $OC(O)R'$, $OC(O)N(R^2)_2$, $-N(R^4)(R^5)$,
 5 $C(O)N(R^5)(R^2)$, $-C(O)R^5$, $-N(R^2)C(O)N(R^2)(R^5)$, $-NC(O)OR^5$, $-OC(O)N(R^2)(R^5)$, or -J; a 5-6 membered carbocyclic or heterocyclic ring system optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, or $SO_2N(R^2)_2$; or a 8-10 membered carbocyclic or heterocyclic ring system
 10 optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, or $SO_2N(R^2)_2$; provided that W is not an R^3 substituted C_1 alkyl.

W' is selected from $N(R^2)-SO_2-Q_2$; $N(R^2)-CO_2-Q_2$; $N(R^2)-C(O)-Q_2$; $N(R^2)(Q_2)$; $C(O)-Q_2$; CO_2-Q_2 ; $C(O)N(R^2)(Q_2)$;
 15 $C(R^2)_2Q_2$.

Each R is independently selected from hydrogen, $-R^2$, $-N(R^2)_2$, $-OR^2$, SR^2 , $-C(O)-N(R^2)_2$, $-S(O_2)-N(R^2)_2$, $-C(O)-OR^2$ or $-C(O)R^2$ wherein two adjacent R are optionally bound to one another and, together with each Y to which
 20 they are respectively bound, form a 4-8 membered carbocyclic or heterocyclic ring.

R^2 is selected from hydrogen, (C_1-C_3) -alkyl, or (C_1-C_3) -alkenyl; each optionally substituted with $-N(R')_2$, $-OR'$, SR' , $-C(O)-N(R')_2$, $-S(O_2)-N(R')_2$, $-C(O)-OR'$,
 25 $-NSO_2R^4$, $-NSO_2R^3$, $-C(O)N(R')(R^3)$, $-NC(O)R^4$, $-N(R')(R^3)$, $-N(R')(R^4)$, $-C(O)R^3$, $-C(O)N(R')(R^4)$, $-N(R^4)_2$, $-C(O)N=C(NH)_2$ or R^3 .

Y is N or C.

Z is CH, N, $C(OCH_3)$, $C(CH_3)$, $C(NH_2)$, $C(OH)$ or
 30 $C(F)$.

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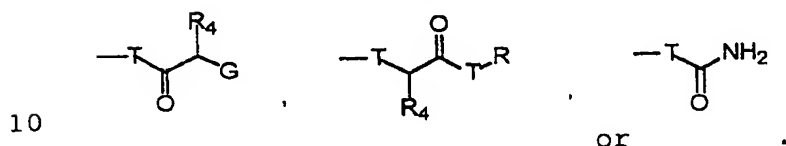
U is selected from R or W.

V is selected from $-C(O)NH_2$, $-P(O)(NH_2)_2$, or $-SO_2NH_2$.

A, B, and C are independently selected from -O-,
5 -CHR'-, -CHR⁴-, -NR'-, -NR⁴- or -S-.

J is a (C₁-C₄) straight chain or branched alkyl derivative substituted with 1-3 substituents selected from D, -T-C(O)R', or -OPO₃H₂.

D is selected from the group



T is either 0 or NH.

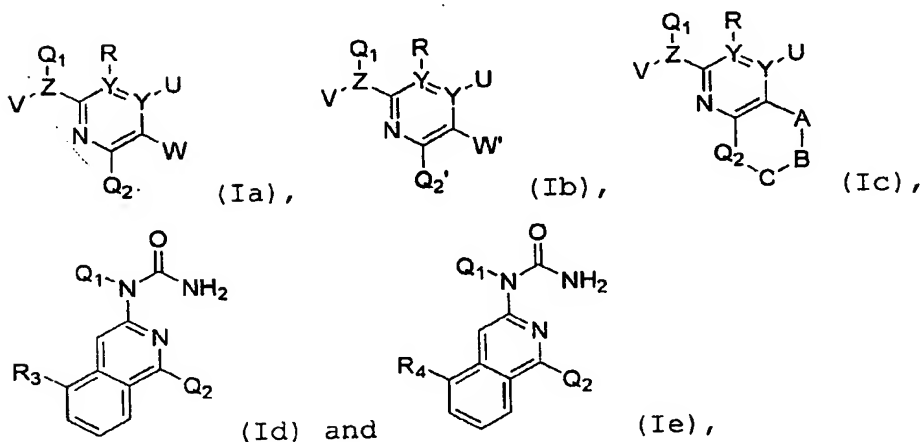
G is either NH_2 or OH .

15 In another embodiment, the invention provides pharmaceutical compositions comprising the p38 inhibitors of this invention. These compositions may be utilized in methods for treating or preventing a variety of disorders, such as cancer, inflammatory diseases, autoimmune diseases, destructive bone disorders, proliferative disorders, infectious diseases, viral diseases and neurodegenerative diseases. These compositions are also useful in methods for preventing cell death and hyperplasia and therefore may be used to treat or prevent reperfusion/ischemia in stroke, heart attacks, and organ hypoxia. The compositions are also useful in methods for preventing thrombin-induced platelet aggregation. Each of these above-described methods is also part of the present invention.

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DETAILED DESCRIPTION OF THE INVENTION

These compounds have the general formula:



wherein each of Q_1 and Q_2 are independently selected from
 5 a phenyl or 5-6 membered aromatic heterocyclic ring
 system, or a 8-10 membered bicyclic ring system
 comprising aromatic carbocyclic rings, aromatic
 heterocyclic rings or a combination of an aromatic
 carbocyclic ring and an aromatic heterocyclic ring.

10 The rings that make up Q_1 are substituted with
 1 to 4 substituents, each of which is independently
 selected from halo; C_1 - C_3 alkyl optionally substituted
 with NR'_2 , OR' , CO_2R' or $CONR'_2$; O -(C_1 - C_3)-alkyl
 optionally substituted with NR'_2 , OR' , CO_2R' or $CONR'_2$;
 15 NR'_2 ; OCF_3 ; CF_3 ; NO_2 ; CO_2R' ; $CONR'$; SR' ; $S(O_2)N(R')_2$; SCF_3 ;
 CN ; $N(R')C(O)R^4$; $N(R')C(O)OR^4$; $N(R')C(O)C(O)R^4$;
 $N(R')S(O_2)R^4$; $N(R')R^4$; $N(R^4)_2$; OR^4 ; $OC(O)R^4$; $OP(O)_3H_2$; or
 $N=C-N(R')_2$.

The rings that make up Q_2 are optionally
 20 substituted with up to 4 substituents, each of which is
 independently selected from halo; C_1 - C_3 straight or -

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branched alkyl optionally substituted with NR'_2 , OR' , $\text{CO}_2\text{R}'$, $\text{S}(\text{O}_2)\text{N}(\text{R}')_2$, $\text{N}=\text{C}-\text{N}(\text{R}')_2$, R^3 , or CONR'_2 ; $\text{O}-(\text{C}_1-\text{C}_3)$ -alkyl; $\text{O}-(\text{C}_1-\text{C}_3)$ -alkyl optionally substituted with NR'_2 , OR' , $\text{CO}_2\text{R}'$, $\text{S}(\text{O}_2)\text{N}(\text{R}')_2$, $\text{N}=\text{C}-\text{N}(\text{R}')_2$, R^3 , or CONR'_2 ; NR'_2 ; OCF_3 ; CF_3 ; NO_2 ; $\text{CO}_2\text{R}'$; CONR' ; R^3 ; OR^3 ; NR^3 ; SR^3 ; $\text{C}(\text{O})\text{R}^3$; $\text{C}(\text{O})\text{N}(\text{R}')\text{R}^3$; $\text{C}(\text{O})\text{OR}^3$; SR' ; $\text{S}(\text{O}_2)\text{N}(\text{R}')_2$; SCF_3 ; $\text{N}=\text{C}-\text{N}(\text{R}')_2$; or CN .

Q_2' is selected from phenyl or a 5-6 member aromatic heterocyclic ring optionally substituted with 1-3 substituents, each of which is independently selected from halogen; C_1-C_3 alkyl optionally substituted with NR'_2 , OR' , $\text{CO}_2\text{R}'$, CONR'_2 , or $\text{O}-\text{P}(\text{O}_3)\text{H}_2$; $\text{O}-(\text{C}_2-\text{C}_3)$ -alkyl optionally substituted with NR'_2 , OR' , $\text{CO}_2\text{R}'$, CONR'_2 , or $\text{OP}(\text{O}_3)\text{H}_2$; OCF_3 ; CF_3 ; OR^4 ; $\text{O}-\text{CO}_2\text{R}^4$; $\text{O}-\text{P}(\text{O}_3)\text{H}_2$; $\text{CO}_2\text{R}'$; CONR' ; SR' ; $\text{S}(\text{O}_2)\text{N}(\text{R}')_2$; SCF_3 ; CN ; $\text{N}(\text{R}')\text{C}(\text{O})\text{R}^4$; $\text{N}(\text{R}')\text{C}(\text{O})\text{OR}^4$; $\text{N}(\text{R}')\text{C}(\text{O})\text{C}(\text{O})\text{R}^4$; $\text{N}(\text{R}')\text{S}(\text{O}_2)\text{R}^4$; $\text{N}(\text{R}')\text{R}^4$; $\text{N}(\text{R}^4)_2$; OR^4 ; $\text{OC}(\text{O})\text{R}^4$; $\text{OP}(\text{O})_3\text{H}_2$; or $\text{N}=\text{C}-\text{N}(\text{R}')_2$; provided that Q_2' is not phenyl optionally substituted 1 to 3 substituents independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl.

R' is selected from hydrogen; (C_1-C_3) -alkyl; (C_2-C_3) -alkenyl or alkynyl; phenyl or phenyl substituted with 1 to 3 substituents independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl; or a 5-6 membered heterocyclic ring system optionally substituted with 1 to 3 substituents independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl.

R^3 is selected from 5-8 membered aromatic or non-aromatic carbocyclic or heterocyclic ring systems

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each optionally substituted with R' , R^4 , $-C(O)R'$, $-C(O)R^4$, $-C(O)OR^4$ or $-J$; or an 8-10 membered bicyclic ring system comprising aromatic carbocyclic rings, aromatic heterocyclic rings or a combination of an aromatic carbocyclic ring and an aromatic heterocyclic ring each optionally substituted with R' , R^4 , $-C(O)R'$, $-C(O)R^4$, $-C(O)OR^4$ or $-J$.

R^4 is (C_1-C_4) -straight or branched alkyl optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, or $SO_2N(R^2)_2$; or a 5-6 membered carbocyclic or heterocyclic ring system optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, or $SO_2N(R^2)_2$.

R^5 is selected from hydrogen; (C_1-C_3) -alkyl optionally substituted with R^3 ; (C_2-C_3) -alkenyl or alkynyl each optionally substituted with R^3 ; phenyl or phenyl substituted with 1 to 3 substituents independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl; or a 5-6 membered heterocyclic ring system optionally substituted with 1 to 3 substituents independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl.

W is selected from $N(R^2)SO_2-N(R^2)_2$; $N(R^2)SO_2-N(R^2)(R^3)$; $N(R^2)C(O)-OR^2$; $N(R^2)C(O)-N(R^2)_2$; $N(R^2)C(O)-N(R^2)(R^3)$; $N(R^2)C(O)-R^2$; $N(R^2)_2$; $C(O)-R^2$; $CH(OH)-R^2$; $C(O)-N(R^2)_2$; $C(O)-OR^2$; J ; or (C_1-C_4) straight or branched alkyl optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, R^3 , $SO_2N(R^2)_2$, $OC(O)R^2$, $OC(O)R'$, $OC(O)N(R^2)_2$, $-N(R^4)(R^5)$, $-C(O)N(R^5)(R^2)$, $-C(O)R^5$, $-N(R^2)C(O)N(R^2)(R^5)$, $-NC(O)OR^5$, $-OC(O)N(R^2)(R^5)$, or $-J$; a 5-6 membered carbocyclic or heterocyclic ring system optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, or $SO_2N(R^2)_2$; or a 8-10

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membered carbocyclic or heterocyclic ring system optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, or $SO_2N(R^2)_2$; provided that W is not an R^3 substituted C_1 alkyl.

5 W' is selected from $N(R^2)-SO_2-Q_2$; $N(R^2)-CO_2-Q_2$; $N(R^2)-C(O)-Q_2$; $N(R^2)(Q_2)$; $C(O)-Q_2$; CO_2-Q_2 ; $C(O)N(R^2)(Q_2)$; $C(R^2)_2Q_2$.

Each R is independently selected from hydrogen, $-R^2$, $-N(R^2)_2$, $-OR^2$, SR^2 , $-C(O)-N(R^2)_2$, $-S(O_2)-N(R^2)_2$,
10 $-C(O)-OR^2$ or $-C(O)R^2$ wherein two adjacent R are optionally bound to one another and, together with each Y to which they are respectively bound, form a 4-8 membered carbocyclic or heterocyclic ring.

When the two R components form a ring together
15 with the Y components to which they are respectively bound, it will be obvious to those skilled in the art that a terminal hydrogen from each unfused R component will be lost. For example, if a ring structure is formed by binding those two R components together, one being $-NH-$
20 CH_3 and the other being $-CH_2-CH_3$, one terminal hydrogen on each R component (indicated in bold) will be lost. Therefore, the resulting portion of the ring structure will have the formula $-NH-CH_2-CH_2-CH_2-$.

R^2 is selected from hydrogen, (C_1-C_3) -alkyl, or
25 (C_1-C_3) -alkenyl; each optionally substituted with $-N(R')_2$, $-OR'$, SR' , $-C(O)-N(R')_2$, $-S(O_2)-N(R')_2$, $-C(O)-OR'$, $-NSO_2R^4$, $-NSO_2R^3$, $-C(O)N(R')(R^3)$, $-NC(O)R^4$, $-N(R')(R^3)$, $-N(R')(R^4)$, $-C(O)R^3$, $-C(O)N(R')(R^4)$, $-N(R^4)_2$, $-C(O)N=C(NH)_2$ or R^3 .

30 Y is N or C.

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Z is CH, N, C(OCH₃), C(CH₃), C(NH₂), C(OH) or C(F).

U is selected from R or W.

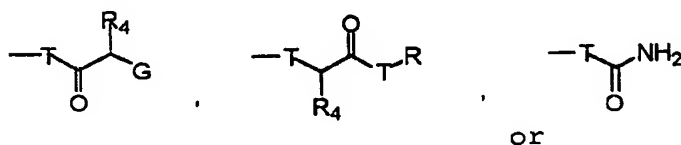
V is selected from -C(O)NH₂, -P(O)(NH₂)₂, or

5 -SO₂NH₂.

A, B, and C are independently selected from -O-, -CHR'-, -CHR⁴-, -NR'-, -NR⁴- or -S-.

J is a (C₁-C₄) straight chain or branched alkyl derivative substituted with 1-3 substituents selected
10 from D, -T-C(O)R', or -OPO₃H₂.

D is selected from the group



15 T is either O or NH.

G is either NH₂ or OH.

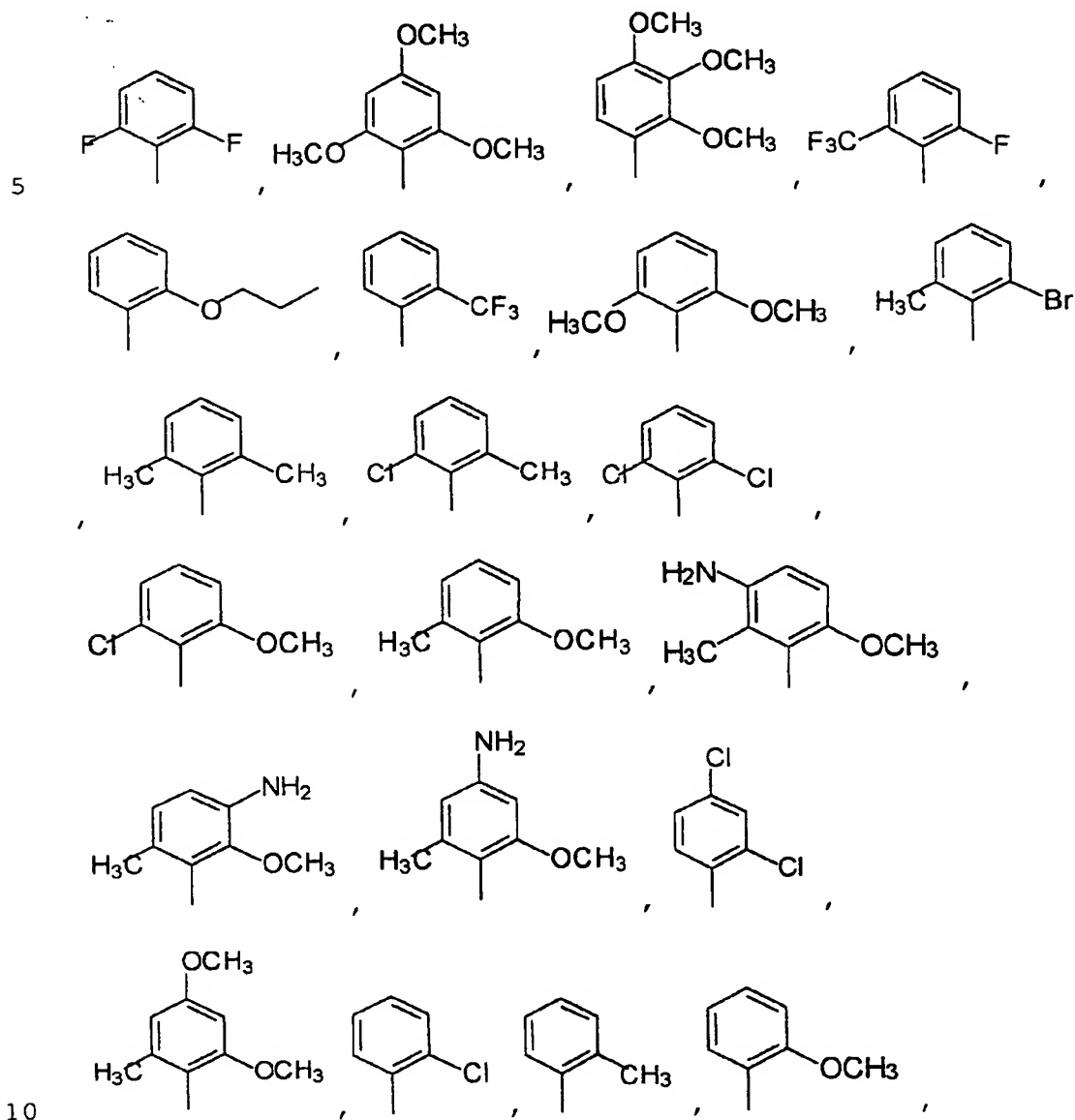
According to a preferred embodiment, Q₁ is selected from phenyl or pyridyl containing 1 to 3 substituents, wherein at least one of said substituents
20 is in the ortho position and said substituents are independently selected from chloro, fluoro, bromo, -CH₃, -OCH₃, -OH, -CF₃, -OCF₃, -O(CH₂)₂CH₃, NH₂, 3,4-methylenedioxy, -N(CH₃)₂, -NH-S(O)₂-phenyl, -NH-C(O)O-CH₂-4-pyridine, -NH-C(O)CH₂-morpholine, -NH-C(O)CH₂-N(CH₃)₂,
25 -NH-C(O)CH₂-piperazine, -NH-C(O)CH₂-pyrrolidine, -NH-C(O)C(O)-morpholine, -NH-C(O)C(O)-piperazine, -NH-C(O)C(O)-pyrrolidine, -O-C(O)CH₂-N(CH₃)₂, or -O-(CH₂)₂-N(CH₃)₂.

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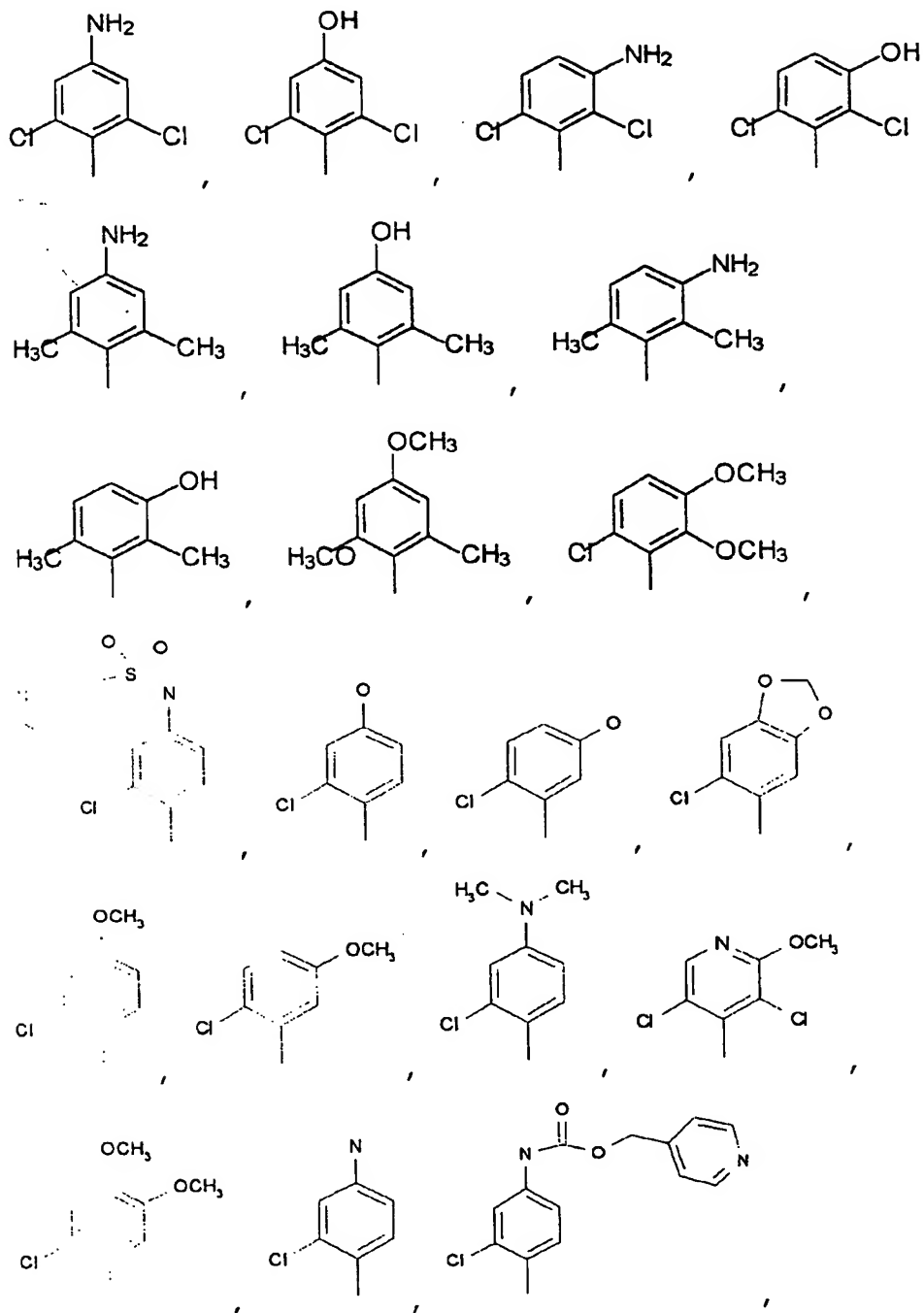
Even more preferred are phenyl or pyridyl containing at least 2 of the above-indicated substituents both being in the ortho position.

Some specific examples of preferred Q₁ are:

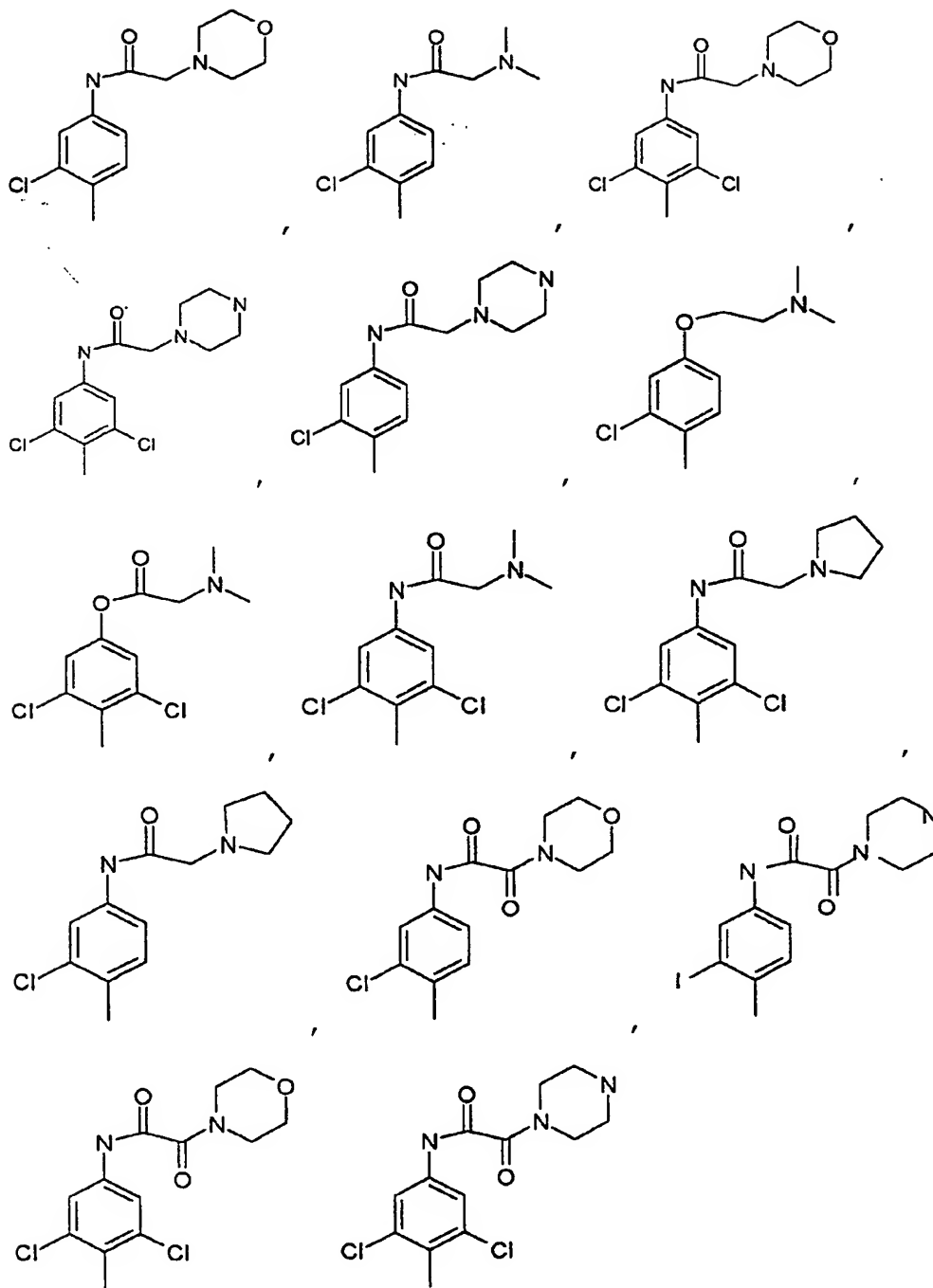


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5

Most preferably, Q₁ is selected from 2-fluoro-6-trifluoromethylphenyl, 2,6-difluorophenyl, 2,6-

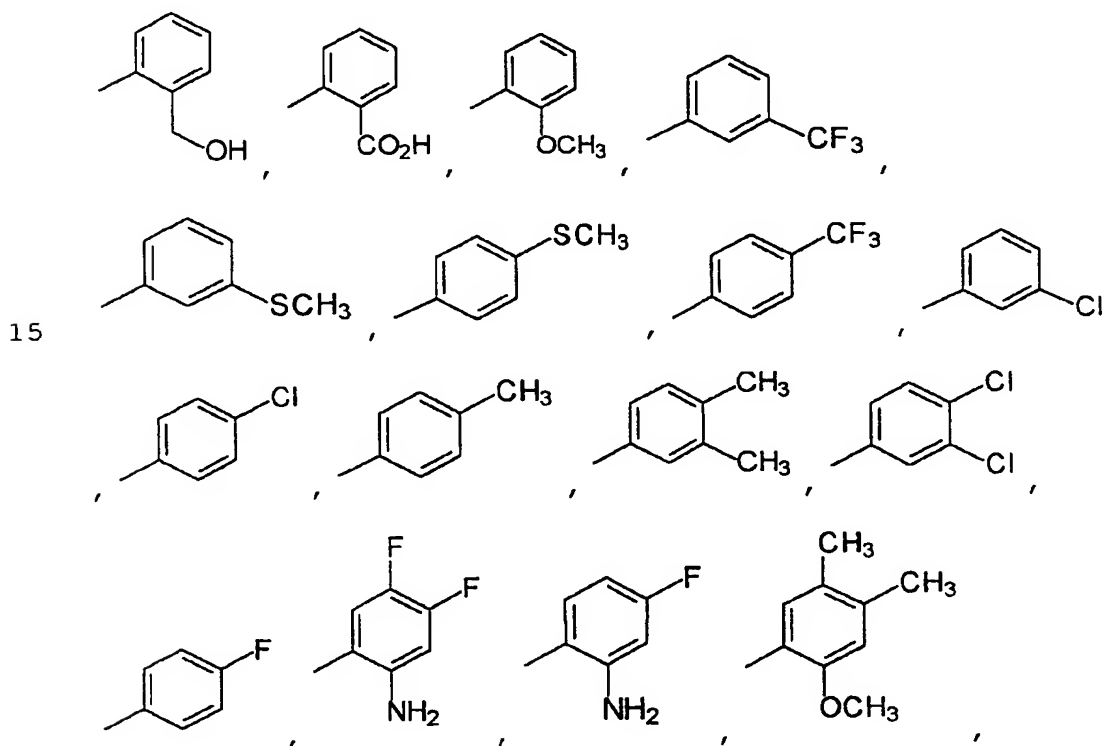
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dichlorophenyl, 2-chloro-4-hydroxyphenyl, 2-chloro-4-aminophenyl, 2,6-dichloro-4-aminophenyl, 2,6-dichloro-3-aminophenyl, 2,6-dimethyl-4-hydroxyphenyl, 2-methoxy-3,5-dichloro-4-pyridyl, 2-chloro-4,5 methylenedioxy phenyl,
 5 or 2-chloro-4-(N-2-morpholino-acetamido)phenyl.

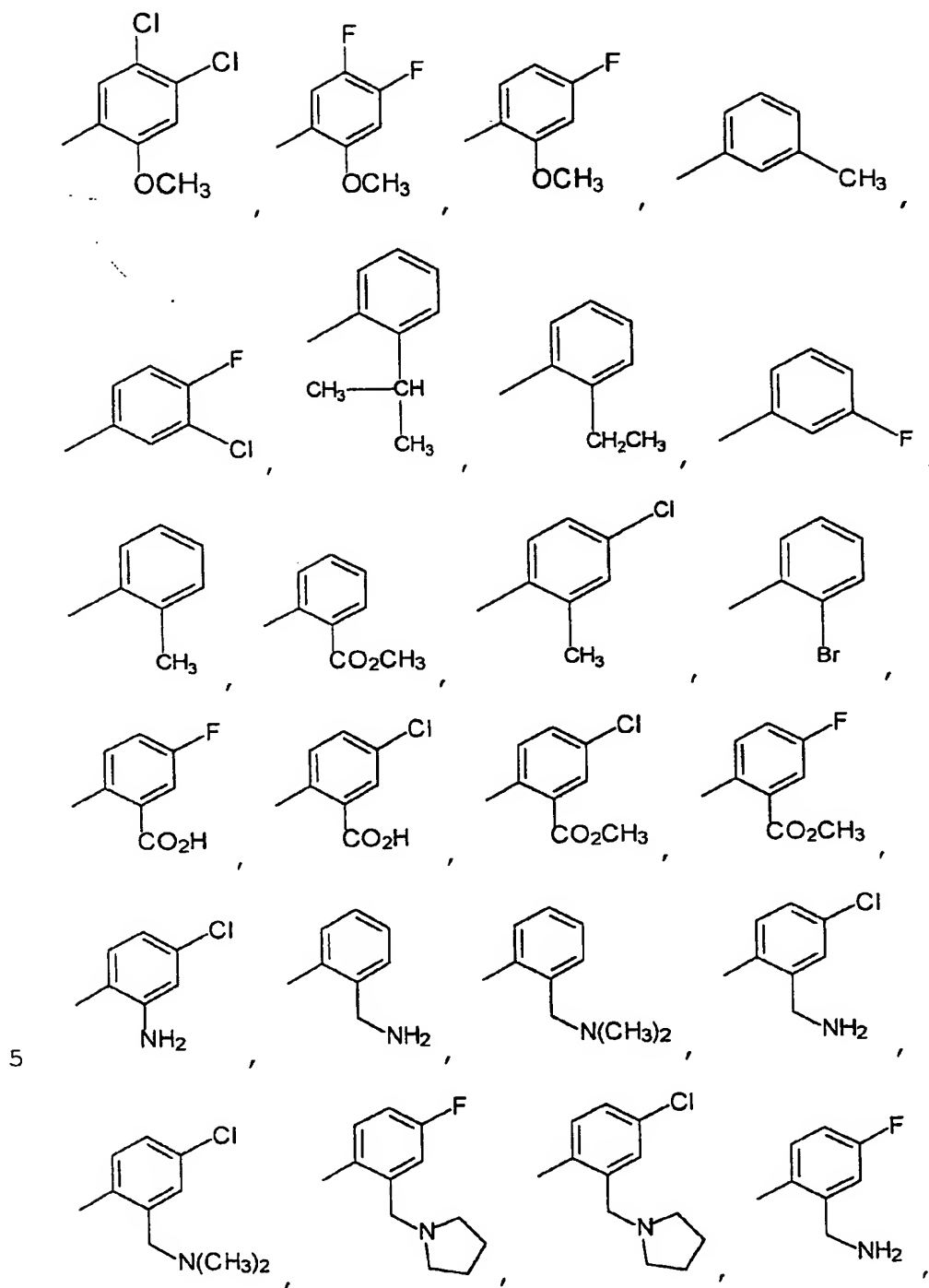
According to a preferred embodiment, Q₂ is phenyl, pyridyl or naphthyl containing 0 to 3 substituents, wherein each substituent is independently selected from chloro, fluoro, bromo, methyl, ethyl,
 10 isopropyl, -OCH₃, -OH, -NH₂, -CF₃, -OCF₃, -SCH₃, -OCH₃, -C(O)OH, -C(O)OCH₃, -CH₂NH₂, -N(CH₃)₂, -CH₂-pyrrolidine and -CH₂OH.

Some specific examples of preferred Q₂ are:

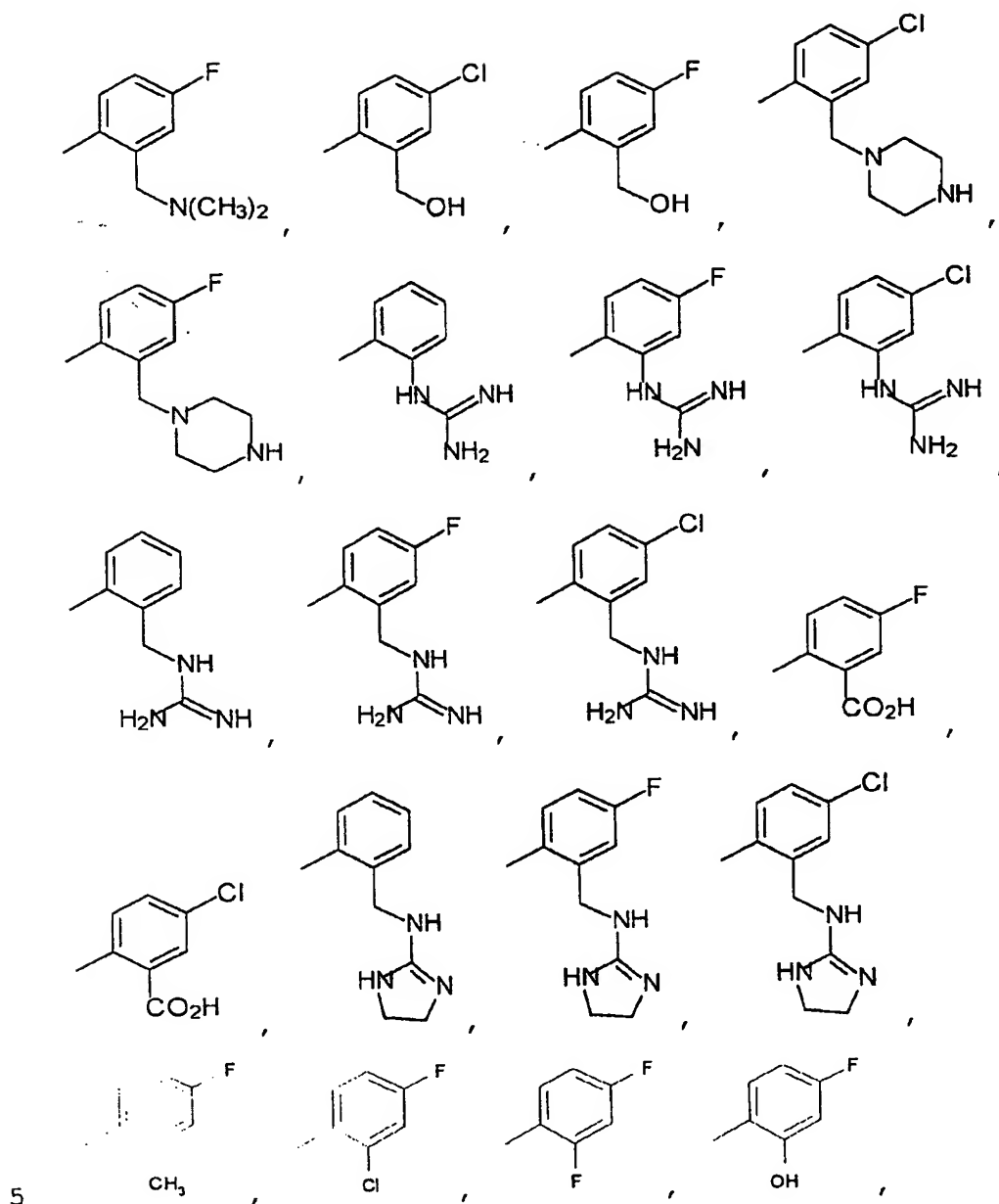


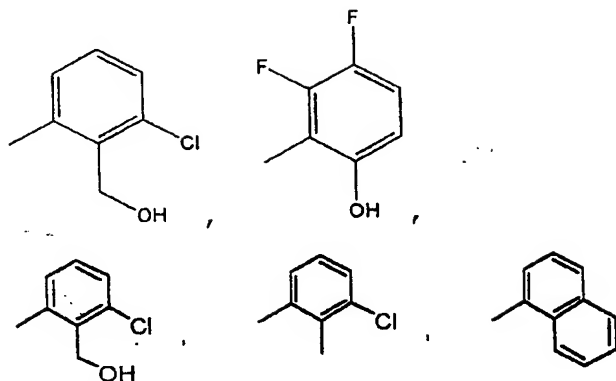
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unsubstituted 2-pyridyl or unsubstituted phenyl.

Most preferred are compounds wherein Q₂ is
 5 selected from phenyl, 2-isopropylphenyl, 3,4-
 dimethylphenyl, 2-ethylphenyl, 3-fluorophenyl, 2-
 methylphenyl, 3-chloro-4-fluorophenyl, 3-chlorophenyl, 2-
 carbomethoxyphenyl, 2-carboxyphenyl, 2-methyl-4-
 chlorophenyl, 2-bromophenyl, 2-pyridyl, 2-
 10 methylenedioxyphenyl, 4-fluorophenyl, 2-methyl-4-
 fluorophenyl, 2-chloro-4-fluorophenyl, 2,4-difluorophenyl,
 2-hydroxy-4-fluorophenyl, 2-methylenedioxy-4-
 fluorophenyl, 1-naphthyl, 3-chloro-2-methylenedioxy, 3-
 chloro-2-methyl, or 4-fluoro-2-methyl.

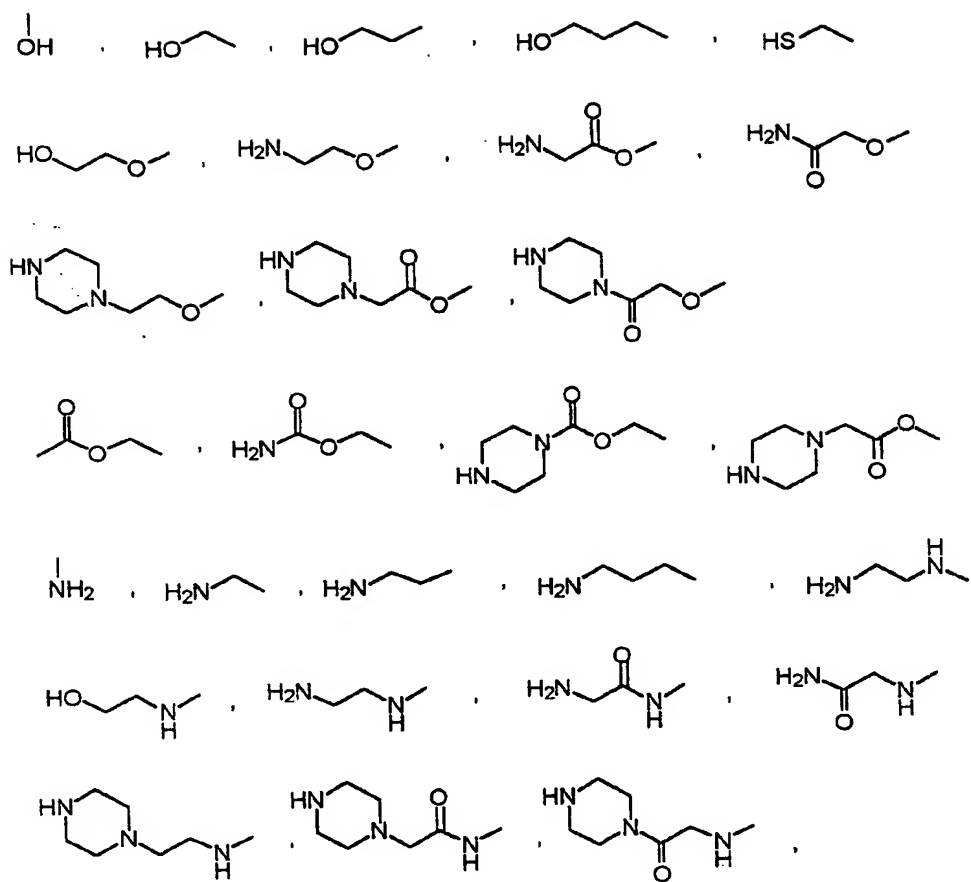
15 According to another preferred embodiment, each
 Y is C.

According to an even more preferred embodiment,
 each Y is C and the R and U attached to each Y component
 is selected from hydrogen or methyl.

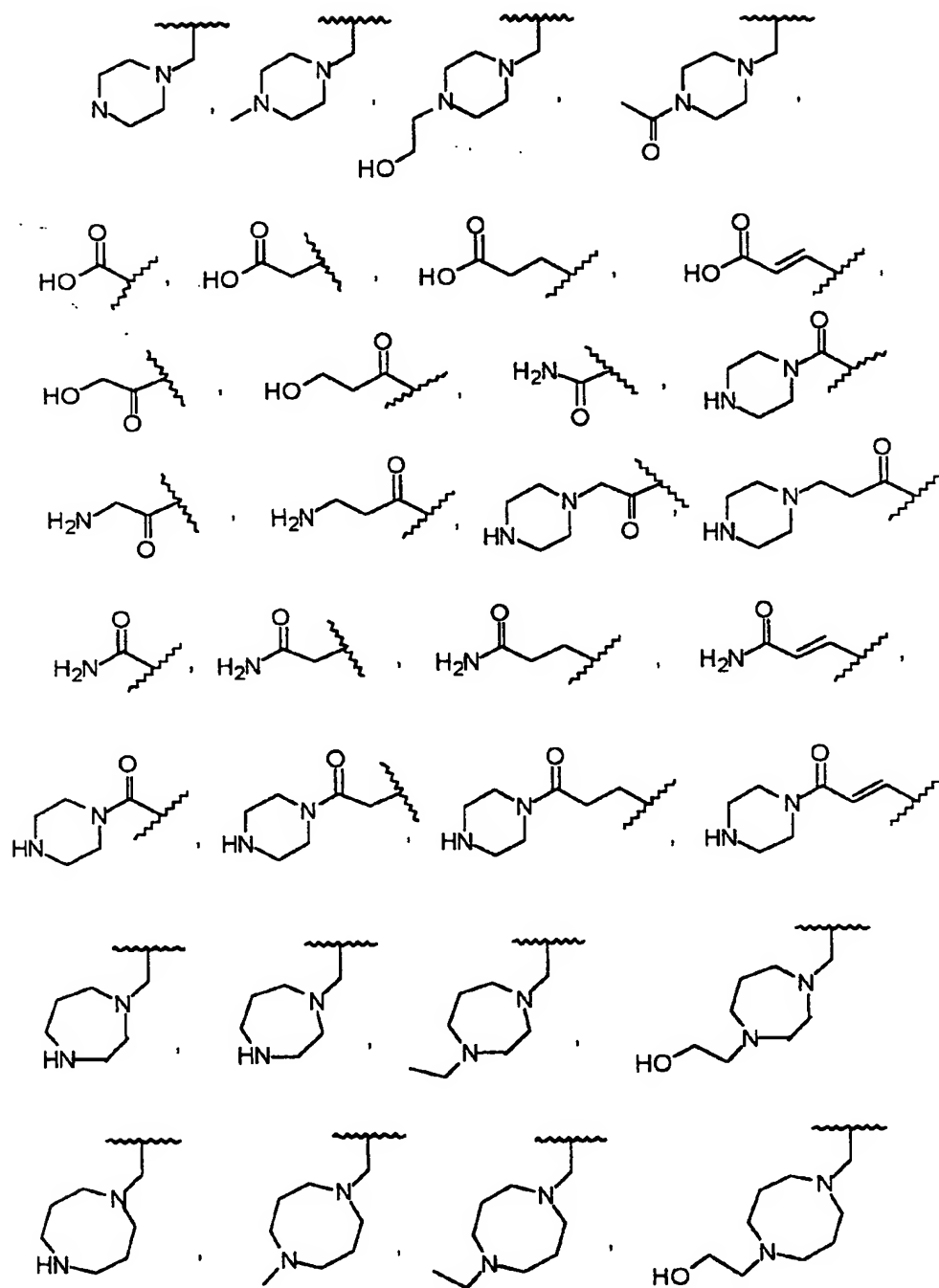
20 According to another preferred embodiment, W is
 a 0-4 atom chain terminating in an alcohol, amine,
 carboxylic acid, ester, amide, or heterocycle.

Some specific examples of preferred W are:

-20-

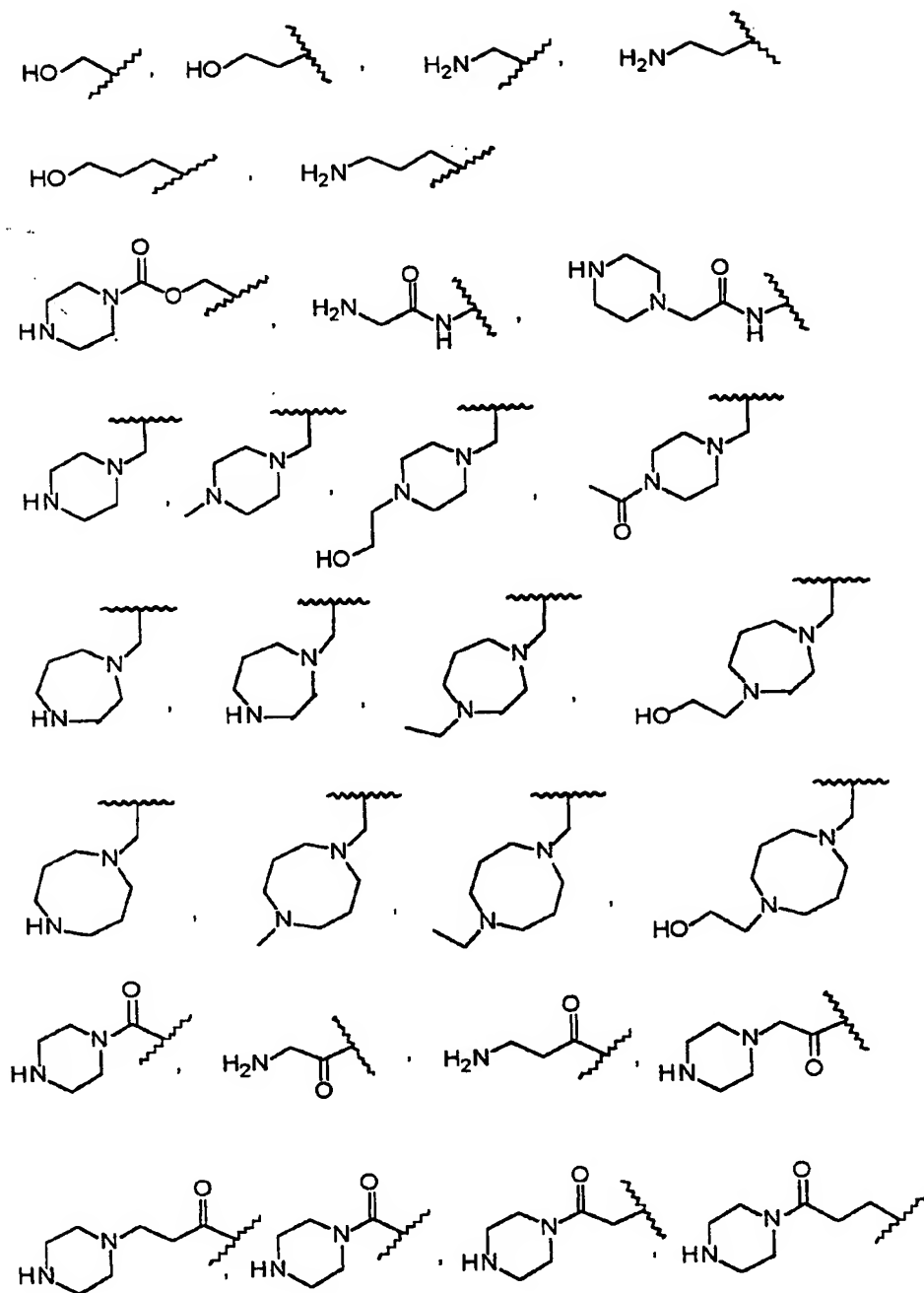
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Most preferably, W is selected from:

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U has the same preferred and most preferred embodiments as W.

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According to an even more preferred embodiment, each Y is C, and W and/or U is not hydrogen.

Some preferred embodiments are provided in Table 1 to 6 below:

Table 1.

Cmpd Number	Structure	Cmpd Number	Structure
VRT-042175		VRT-101262	
VRT-041238		VRT-043176	
VRT-042305		VRT-043180	
VRT-043675		VRT-043181	
VRT-042313		VRT-101259	
VRT-101257		VRT-042196	

Table 2.

Cmpd Number	Structure	Cmpd Number	Structure
VRT- 043188		VRT- 043674	
VRT- 100306		VRT- 100318	
VRT- 043190		VRT- 101256	
VRT- 043192		VRT- 101255	
VRT- 043672		VRT- 101253	
VRT- 043673		VRT- 101251	

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Table 3.

Cmpd Number	Structure	Cmpd Number	Structure
VRT-100325		VRT-100324	
VRT-043683		VRT-101249	
VRT-101248		VRT-043693	
VRT-043685		VRT-043694	
VRT-043686		VRT-043695	
VRT-043690		VRT-101247	

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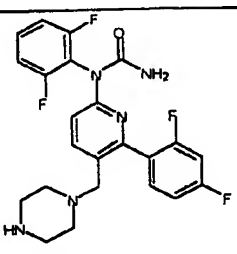
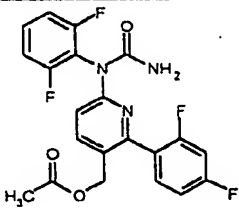
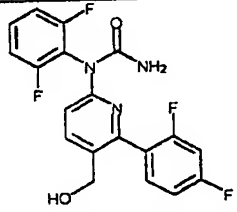
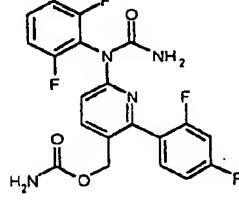
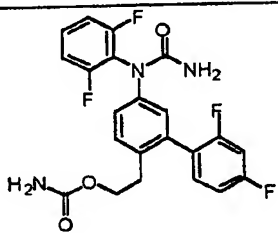
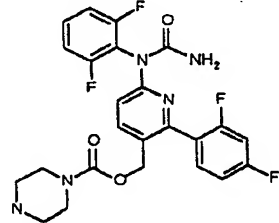
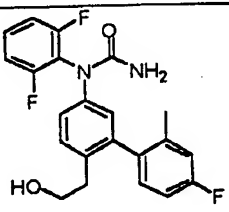
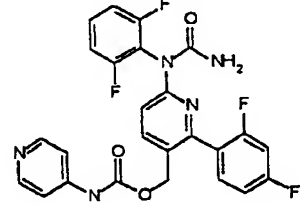
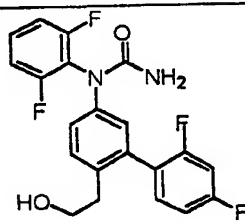
Table 4.

Cmpd Number	Structure	Cmpd Number	Structure
VRT-100310		VRT-043688	
VRT-043678		VRT-043675	
VRT-043191		VRT-042307	
VRT-100019		VRT-040569	
VRT-100020		VRT-100025	

Table 5.

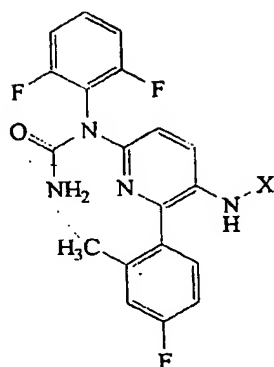
Cmpd Number	Structure	Cmpd Number	Structure
VRT-100026		VRT-100313	
VRT-100304		VRT-100315	
VRT-100305		VRT-100317	
VRT-041291		VRT-37742	
VRT-032884		VRT-100323	
VRT-034465		VRT-100146	

Table 6.

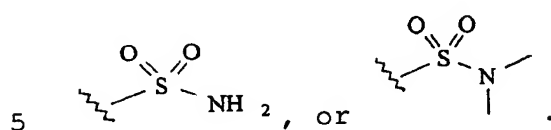
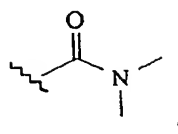
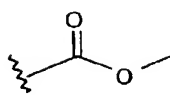
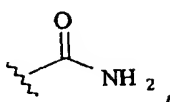
Cmpd Number	Structure	Cmpd Number	Structure
VRT-101094		VRT-043605	
VRT-043631		VRT-100075	
VRT-043008		VRT-100076	
VRT-042266		VRT-100077	
VRT-042169			

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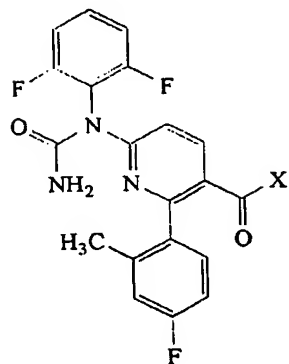
Particularly preferred embodiments include:



wherein X is H,



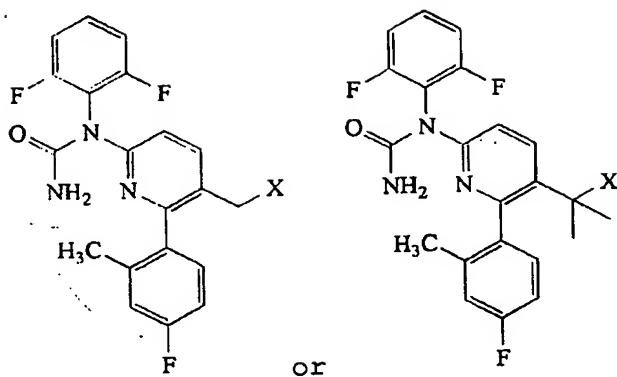
Particularly preferred embodiments also include:



wherein X is NH_2 or $\text{N}(\text{CH}_3)_2$;

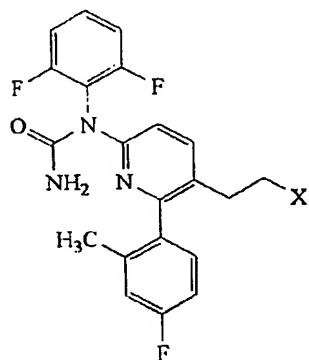
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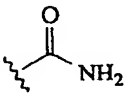
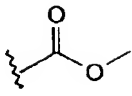


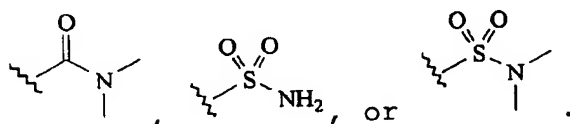
wherein X is OH, NH₂, or N(CH₃)₂.

Other particularly preferred embodiments include:



5

wherein X is OH, NH₂, N(CH₃)₂, , ,

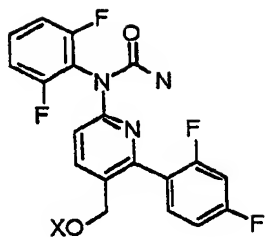


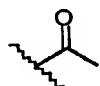
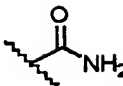
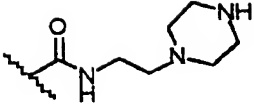
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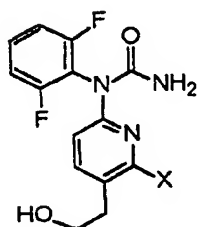
Other particularly preferred embodiments include:

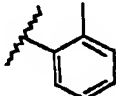
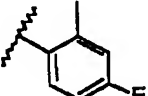
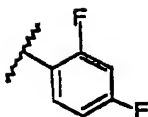
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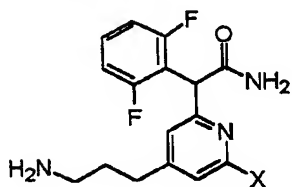


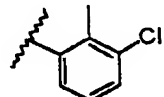
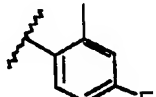
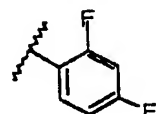
wherein X = H, , ,  and



wherein X = , , .

Other particularly preferred embodiments include:

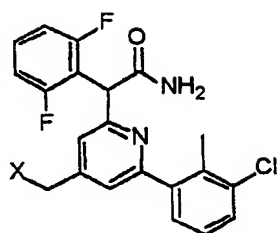


wherein X = , ,  and

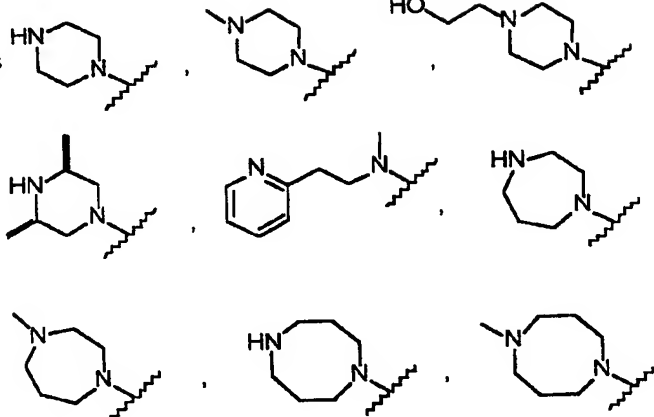
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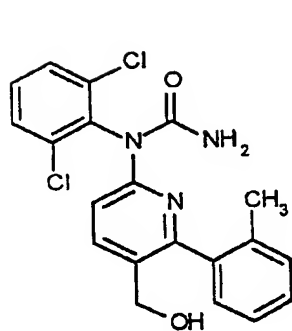
-33-



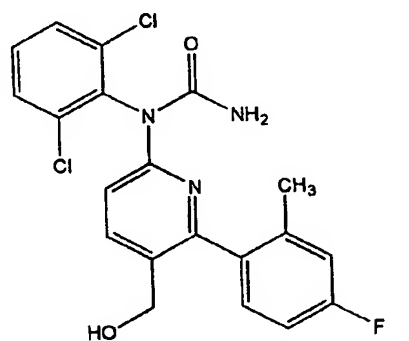
wherein X is



Most preferred embodiments include:



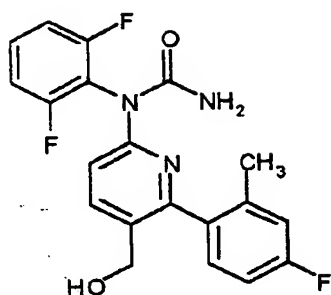
5 (Compound 17)



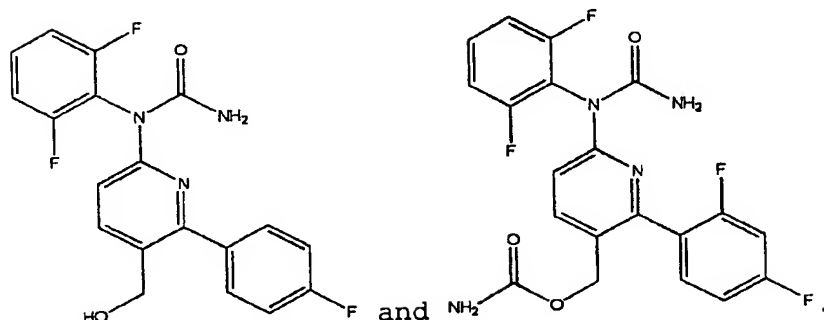
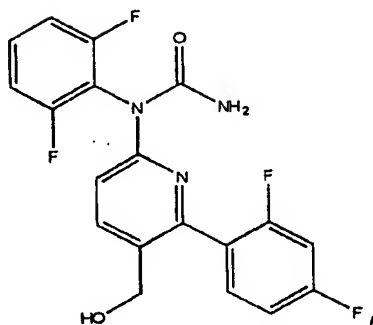
(Compound 18)

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(Compound 19)



5 According to another embodiment, the present invention provides methods of producing the above-identified inhibitors of p38 of the formulae (Ia), (Ib), (Ic), (Id) and (Ie). Representative synthesis schemes for formula (Ia) are depicted below.

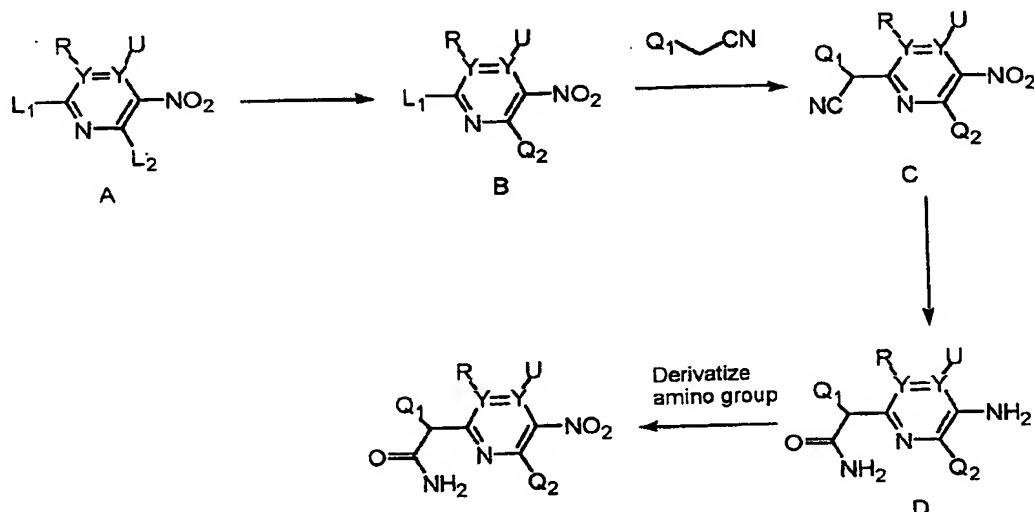
10 Schemes 1-3 illustrate the preparation of compounds in which W is either an amino, carboxyl or an aldehyde function. In each case the particular moiety may be modified through chemistry well known in the literature. For example the final amino compounds D and

15 N (schemes 1 and 4 respectively) may be acylated, sulfonylated or alkylated to prepare compounds within the scope of W. In all schemes, the L1 and L2 groups on the initial materials are meant to represent leaving groups

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ortho to the nitrogen atom in a heterocyclic ring. For example, compound A may be 2,6-dichloro-3 nitro pyridine.

Scheme 1



5 In Scheme 1, W is selected from amino-derivatized compounds such as $N(R^2)SO_2-N(R^2)_2$; $N(R^2)SO_2-N(R^2)(R^3)$; $N(R^2)C(O)-OR^2$; $N(R^2)C(O)-N(R^2)_2$; $N(R^2)C(O)-N(R^2)(R^3)$; $N(R^2)C(O)-R^2$; or $N(R^2)_2$.

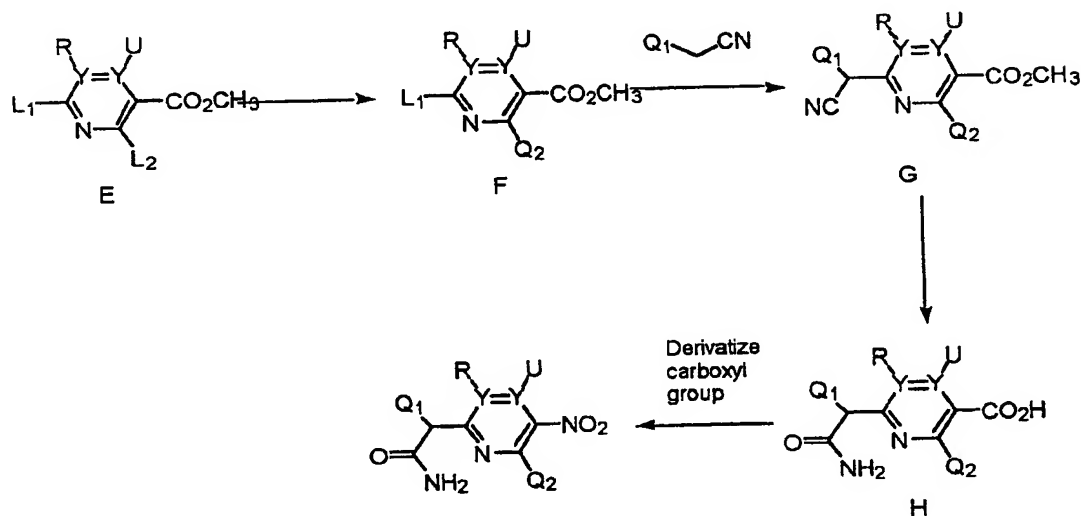
10 In Scheme 1, the Q_2 ring is introduced utilizing one of many reactions known in the art which result in the production of biaryl compounds. One example may be the reaction of an aryl lithium compound with the pyridine intermediate A. Alternatively, an arylmetallic compound such as an aryl stannane or an aryl
15 boronic acid may be reacted with the aryl halide portion (intermediate A) in the presence of a Pd^0 catalyst to form product B. In the next step, a Q_1 substituted derivative such as a phenyl acetonitrile derivative may be treated with a base such as sodium hydride, sodium
20 amide, LDA, lithium hexamethyldisilazide or any number of

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other non-nucleophilic bases to deprotonate the position alpha to the cyano group, which represents a masked amide moiety. This anion is then contacted with intermediate B to form C. The nitrile or equivalent group of

5 intermediate C is then hydrolyzed to form the amide and the nitro group is subjected to reducing conditions to form the amine intermediate D. Intermediate D is then used to introduce various functionality defined by W through chemistry such as acylation, sulfonylation or

10 alkylation reactions well known in the literature. Depending on the regiochemistry of the first two steps of this procedure, the first two steps may need to be reversed.

15 Scheme 2

In Scheme 2, W is selected from carboxyl-derivatized compounds such as $C(O)-R^2$; $CH(OH)-R^2$; $C(O)-N(R^2)_2$; or $C(O)-$

20 OR^2 .

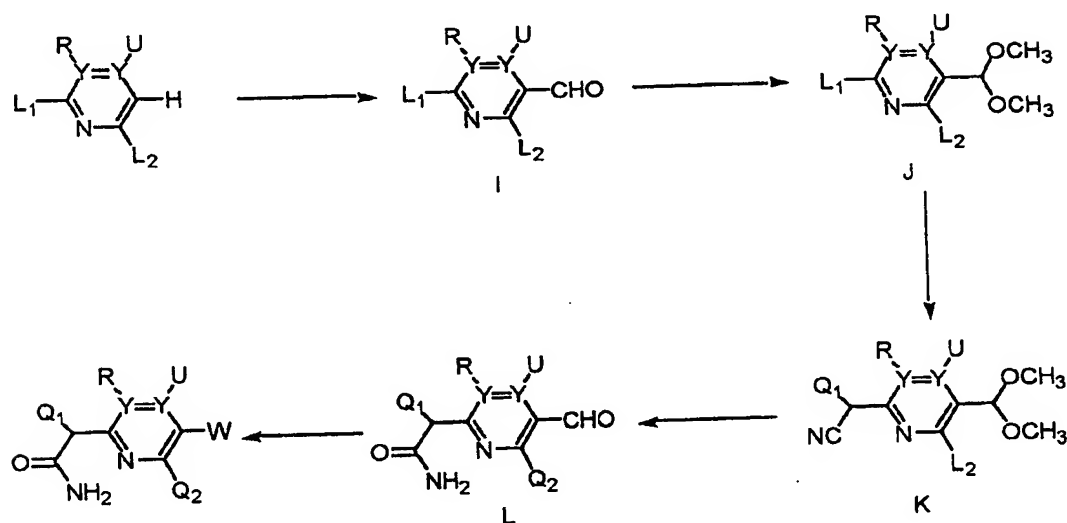
Scheme 2 generally follows the procedures described for Scheme 1 except that a carboxyl

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intermediate such as E is the starting material. The first two steps mirror Scheme 1, and, as mentioned for Scheme 1, may be reversed depending on the regiochemistry of specific examples. Intermediate G is formed from these first two steps and this material may be hydrolyzed as mention to for the carboxyl intermediate H. The carboxyl group may then be modified according to well-known procedures from the literature to prepare analogs with defined W substituents such as acylations, amidations and esterifications.

Scheme 3



In Scheme 3, W is selected from (C₁-C₄) straight or branched alkyl optionally substituted with N(R')₂, OR', CO₂R', CON(R')₂, R³, or SO₂N(R²)₂; or a 5-6 membered carbocyclic or heterocyclic ring system optionally substituted with N(R')₂, OR', CO₂R', CON(R')₂, or SO₂N(R²)₂; provided that W is not an R³ substituted C₁ alkyl.

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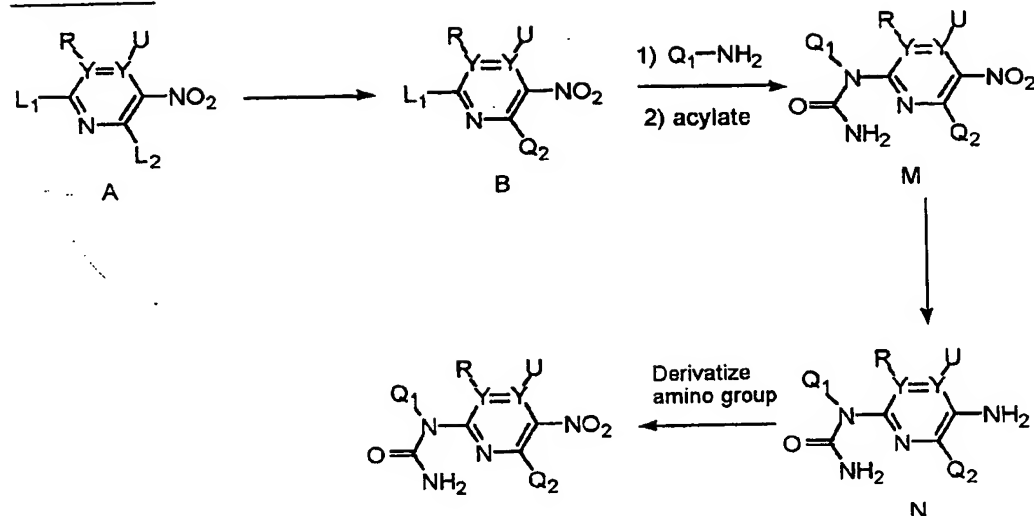
In scheme 3 a pyridine derivative is metalated and quenched with one of many known electrophiles which can generate an aldehyde, to form intermediate I. The aldehyde can then be masked to form the dimethyl acetal J. This intermediate is then carried on as described in scheme 1 and 2 to introduce the Q1 and Q2 substituents, to produce intermediate L. As before, these two steps may be interchanged depending on specific regiochemistry. The masked aldehyde of L may then be deprotected and utilized to form compounds with the defined W substitution using well know chemistry such as alkylations and reductive aminations.

Schemes 4-6 are similar to schemes 1-3 with the exception that the targeted compounds are those in which Z = Nitrogen. The steps for these schemes parallel 1-3 with the exception that the alkylation utilizing a phenyl acetonitrile is replaced with a reaction with a Q1 amine derivative such as a substituted aniline derivative. The amide portion of the molecule is then introduced in an acylation reaction with, for example, chlorosulfonyl isocyanate.

25

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Scheme 4

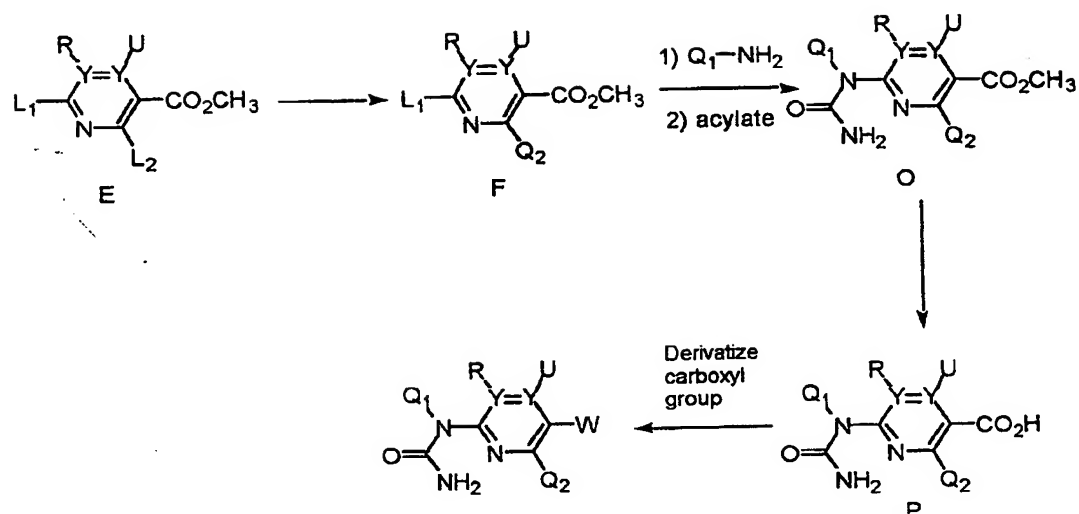


In Scheme 4, W is selected from amino-derivatized groups such as $N(R^2)SO_2-N(R^2)_2$; $N(R^2)SO_2-N(R^2)(R^3)$; $N(R^2)C(O)-OR^2$; $N(R^2)C(O)-N(R^2)_2$; $N(R^2)C(O)-N(R^2)(R^3)$; $N(R^2)C(O)-R^2$; or $N(R^2)_2$.

In Scheme 4, intermediate B (from scheme 1) is treated with, for example, an aniline derivative in the presence of a base such as potassium carbonate. Additionally, a palladium catalyst may be utilized to enhance the reactivity of this general type of reaction, if needed. The resulting amine derivative is then acylated to form intermediate M. The nitro group of M is then reduced to form N and the amino group may then be derivatized as described for scheme 1. As mentioned for schemes 1-3, the steps involved in the introduction of the Q_1 and Q_2 substituents may be interchanged depending on the specific regiochemistry of specific compounds.

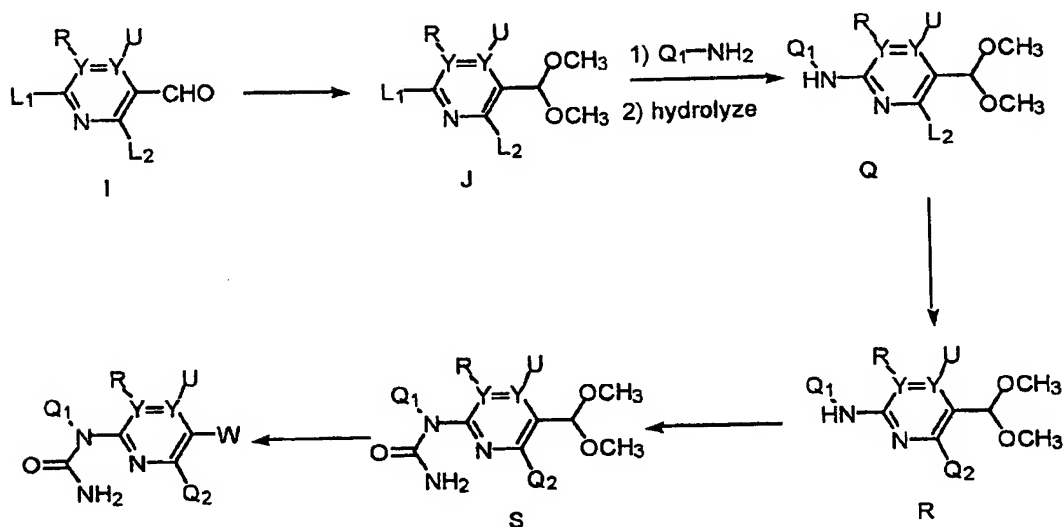
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Scheme 5



In Scheme 5, W is selected from carboxyl-derivatized groups such as $C(O)-R^2$; $CH(OH)-R^2$; $C(O)-N(R^2)_2$; or $C(O)-OR^2$.

Scheme 6



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In Scheme 6, W is selected from (C₁-C₄) straight or branched alkyl optionally substituted with N(R')₂, OR', CO₂R', CON(R')₂, R³, or SO₂N(R²)₂; or a 5-6 membered carbocyclic or heterocyclic ring system optionally substituted with N(R')₂, OR', CO₂R', CON(R')₂, or SO₂N(R²)₂; provided that W is not an R³ substituted C₁ alkyl.

Schemes 5 and 6 generally follow the procedures mentioned above.

One having skill in the art will recognize schemes 1-6 may be used to synthesize compounds having the general formula of (Ib), (Ic), (Id) and (Ie).

According to another embodiment of the invention, the activity of the p38 inhibitors of this invention may be assayed *in vitro*, *in vivo* or in a cell line. *In vitro* assays include assays that determine inhibition of either the kinase activity or ATPase activity of activated p38. Alternate *in vitro* assays quantitate the ability of the inhibitor to bind to p38 and may be measured either by radiolabelling the inhibitor prior to binding, isolating the inhibitor/p38 complex and determining the amount of radiolabel bound, or by running a competition experiment where new inhibitors are incubated with p38 bound to known radioligands.

Cell culture assays of the inhibitory effect of the compounds of this invention may determine the amounts of TNF, IL-1, IL-6 or IL-8 produced in whole blood or cell fractions thereof in cells treated with inhibitor as compared to cells treated with negative controls. Level of these cytokines may be determined through the use of commercially available ELISAs.

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An *in vivo* assay useful for determining the inhibitory activity of the p38 inhibitors of this invention are the suppression of hind paw edema in rats with *Mycobacterium butyricum*-induced adjuvant arthritis. This is described in J.C. Boehm et al., J. Med. Chem., 39, pp. 3929-37 (1996), the disclosure of which is herein incorporated by reference. The p38 inhibitors of this invention may also be assayed in animal models of arthritis, bone resorption, endotoxin shock and immune function, as described in A. M. Badger et al., J. Pharmacol. Experimental Therapeutics, 279, pp. 1453-61 (1996), the disclosure of which is herein incorporated by reference.

The p38 inhibitors or pharmaceutical salts thereof may be formulated into pharmaceutical compositions for administration to animals or humans. These pharmaceutical compositions, which comprise an amount of p38 inhibitor effective to treat or prevent a p38-mediated condition and a pharmaceutically acceptable carrier, are another embodiment of the present invention.

The term "p38-mediated condition", as used herein means any disease or other deleterious condition in which p38 is known to play a role. This includes conditions known to be caused by IL-1, TNF, IL-6 or IL-8 overproduction. Such conditions include, without limitation, inflammatory diseases, autoimmune diseases, destructive bone disorders, proliferative disorders, infectious diseases, neurodegenerative diseases, allergies, reperfusion/ischemia in stroke, heart attacks, angiogenic disorders, organ hypoxia, vascular hyperplasia, cardiac hypertrophy, thrombin-induced

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platelet aggregation, and conditions associated with prostaglandin endoperoxidase synthase-2.

Inflammatory diseases which may be treated or prevented by the compounds of this invention include, but
5 are not limited to, acute pancreatitis, chronic pancreatitis, asthma, allergies, and adult respiratory distress syndrome.

Autoimmune diseases which may be treated or prevented by the compounds of this invention include, but
10 are not limited to, glomerulonephritis, rheumatoid arthritis, systemic lupus erythematosus, scleroderma, chronic thyroiditis, Graves' disease, autoimmune gastritis, diabetes, autoimmune hemolytic anemia, autoimmune neutropenia, thrombocytopenia, atopic
15 dermatitis, chronic active hepatitis, myasthenia gravis, multiple sclerosis, inflammatory bowel disease, ulcerative colitis, Crohn's disease, psoriasis, or graft vs. host disease.

Destructive bone disorders which may be treated
20 or prevented by the compounds of this invention include, but are not limited to, osteoporosis, osteoarthritis and multiple myeloma-related bone disorder.

Proliferative diseases which may be treated or prevented by the compounds of this invention include, but
25 are not limited to, acute myelogenous leukemia, chronic myelogenous leukemia, metastatic melanoma, Kaposi's sarcoma, and multiple myeloma.

Angiogenic disorders which may be treated or prevented by the compounds of this invention include
30 solid tumors, ocular neovascularization, infantile haemangiomas.

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Infectious diseases which may be treated or prevented by the compounds of this invention include, but are not limited to, sepsis, septic shock, and Shigellosis.

5 Viral diseases which may be treated or prevented by the compounds of this invention include, but are not limited to, acute hepatitis infection (including hepatitis A, hepatitis B and hepatitis C), HIV infection and CMV retinitis.

10 Neurodegenerative diseases which may be treated or prevented by the compounds of this invention include, but are not limited to, Alzheimer's disease, Parkinson's disease, cerebral ischemias or neurodegenerative disease caused by traumatic injury.

15 "p38-mediated conditions" also include ischemia/reperfusion in stroke, heart attacks, myocardial ischemia, organ hypoxia, vascular hyperplasia, cardiac hypertrophy, and thrombin-induced platelet aggregation.

 In addition, p38 inhibitors of the instant
20 invention are also capable of inhibiting the expression of inducible pro-inflammatory proteins such as prostaglandin endoperoxide synthase-2 (PGHS-2), also referred to as cyclooxygenase-2 (COX-2). Therefore, other "p38-mediated conditions" which may be treated by the
25 compounds of this invention include edema, analgesia, fever and pain, such as neuromuscular pain, headache, cancer pain, dental pain and arthritis pain.

 The diseases that may be treated or prevented
by the p38 inhibitors of this invention may also be
30 conveniently grouped by the cytokine (IL-1, TNF, IL-6, IL-8) that is believed to be responsible for the disease.

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- Thus, an IL-1-mediated disease or condition includes rheumatoid arthritis, osteoarthritis, stroke, endotoxemia and/or toxic shock syndrome, inflammatory reaction induced by endotoxin, inflammatory bowel disease, tuberculosis, atherosclerosis, muscle degeneration, cachexia, psoriatic arthritis, Reiter's syndrome, gout, traumatic arthritis, rubella arthritis, acute synovitis, diabetes, pancreatic β -cell disease and Alzheimer's disease.
- 10 TNF-mediated disease or condition includes, rheumatoid arthritis, rheumatoid spondylitis, osteoarthritis, gouty arthritis and other arthritic conditions, sepsis, septic shock, endotoxic shock, gram negative sepsis, toxic shock syndrome, adult respiratory
- 15 distress syndrome, cerebral malaria, chronic pulmonary inflammatory disease, silicosis, pulmonary sarcoisosis, bone resorption diseases, reperfusion injury, graft vs. host reaction, allograft rejections, fever and myalgias due to infection, cachexia secondary to infection, AIDS,
- 20 ARC or malignancy, keloid formation, scar tissue formation, Crohn's disease, ulcerative colitis or pyresis. TNF-mediated diseases also include viral infections, such as HIV, CMV, influenza and herpes; and veterinary viral infections, such as lentivirus
- 25 infections, including, but not limited to equine infectious anemia virus, caprine arthritis virus, visna virus or maedi virus; or retrovirus infections, including feline immunodeficiency virus, bovine immunodeficiency virus, or canine immunodeficiency virus.
- 30 IL-8 mediated disease or condition includes diseases characterized by massive neutrophil infiltration, such as psoriasis, inflammatory bowel -

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disease, asthma, cardiac and renal reperfusion injury, adult respiratory distress syndrome, thrombosis and glomerulonephritis.

In addition, the compounds of this invention may be used topically to treat or prevent conditions caused or exacerbated by IL-1 or TNF. Such conditions include inflamed joints, eczema, psoriasis, inflammatory skin conditions such as sunburn, inflammatory eye conditions such as conjunctivitis, pyresis, pain and other conditions associated with inflammation.

In addition to the compounds of this invention, pharmaceutically acceptable salts of the compounds of this invention may also be employed in compositions to treat or prevent the above-identified disorders.

Pharmaceutically acceptable salts of the compounds of this invention include those derived from pharmaceutically acceptable inorganic and organic acids and bases. Examples of suitable acid salts include acetate, adipate, alginate, aspartate, benzoate, benzenesulfonate, bisulfate, butyrate, citrate, camphorate, camphorsulfonate, cyclopentanepropionate, digluconate, dodecylsulfate, ethanesulfonate, formate, fumarate, glucoheptanoate, glycerophosphate, glycolate, hemisulfate, heptanoate, hexanoate, hydrochloride, hydrobromide, hydroiodide, 2-hydroxyethanesulfonate, lactate, maleate, malonate, methanesulfonate, 2-naphthalenesulfonate, nicotinate, nitrate, oxalate, palmoate, pectinate, persulfate, 3-phenylpropionate, phosphate, picrate, pivalate, propionate, salicylate, succinate, sulfate, tartrate, thiocyanate, tosylate and undecanoate. Other acids, such as oxalic, while not in themselves pharmaceutically acceptable, may be employed

in the preparation of salts useful as intermediates in obtaining the compounds of the invention and their pharmaceutically acceptable acid addition salts.

Salts derived from appropriate bases include alkali metal (e.g., sodium and potassium), alkaline earth metal (e.g., magnesium), ammonium and N-(C1-4 alkyl)4+ salts. This invention also envisions the quaternization of any basic nitrogen-containing groups of the compounds disclosed herein. Water or oil-soluble or dispersible products may be obtained by such quaternization.

Pharmaceutically acceptable carriers that may be used in these pharmaceutical compositions include, but are not limited to, ion exchangers, alumina, aluminum stearate, lecithin, serum proteins, such as human serum albumin, buffer substances such as phosphates, glycine, sorbic acid, potassium sorbate, partial glyceride mixtures of saturated vegetable fatty acids, water, salts or electrolytes, such as protamine sulfate, disodium hydrogen phosphate, potassium hydrogen phosphate, sodium chloride, zinc salts, colloidal silica, magnesium trisilicate, polyvinyl pyrrolidone, cellulose-based substances, polyethylene glycol, sodium carboxymethylcellulose, polyacrylates, waxes, polyethylene-polyoxypropylene-block polymers, polyethylene glycol and wool fat.

The compositions of the present invention may be administered orally, parenterally, by inhalation spray, topically, rectally, nasally, buccally, vaginally or via an implanted reservoir. The term "parenteral" as used herein includes subcutaneous, intravenous, intramuscular, intra-articular, intra-synovial, intrasternal, intrathecal, intrahepatic, intralesional

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and intracranial injection or infusion techniques. Preferably, the compositions are administered orally, intraperitoneally or intravenously.

Sterile injectable forms of the compositions of this invention may be aqueous or oleaginous suspension. These suspensions may be formulated according to techniques known in the art using suitable dispersing or wetting agents and suspending agents. The sterile injectable preparation may also be a sterile injectable solution or suspension in a non-toxic parenterally-acceptable diluent or solvent, for example as a solution in 1,3-butanediol. Among the acceptable vehicles and solvents that may be employed are water, Ringer's solution and isotonic sodium chloride solution. In addition, sterile, fixed oils are conventionally employed as a solvent or suspending medium. For this purpose, any bland fixed oil may be employed including synthetic mono- or di-glycerides. Fatty acids, such as oleic acid and its glyceride derivatives are useful in the preparation of injectables, as are natural pharmaceutically-acceptable oils, such as olive oil or castor oil, especially in their polyoxyethylated versions. These oil solutions or suspensions may also contain a long-chain alcohol diluent or dispersant, such as carboxymethyl cellulose or similar dispersing agents which are commonly used in the formulation of pharmaceutically acceptable dosage forms including emulsions and suspensions. Other commonly used surfactants, such as Tweens, Spans and other emulsifying agents or bioavailability enhancers which are commonly used in the manufacture of pharmaceutically acceptable solid, liquid, or other

dosage forms may also be used for the purposes of formulation.

The pharmaceutical compositions of this invention may be orally administered in any orally acceptable dosage form including, but not limited to, capsules, tablets, aqueous suspensions or solutions. In the case of tablets for oral use, carriers commonly used include lactose and corn starch. Lubricating agents, such as magnesium stearate, are also typically added. For oral administration in a capsule form, useful diluents include lactose and dried cornstarch. When aqueous suspensions are required for oral use, the active ingredient is combined with emulsifying and suspending agents. If desired, certain sweetening, flavoring or coloring agents may also be added.

Alternatively, the pharmaceutical compositions of this invention may be administered in the form of suppositories for rectal administration. These can be prepared by mixing the agent with a suitable non-irritating excipient which is solid at room temperature but liquid at rectal temperature and therefore will melt in the rectum to release the drug. Such materials include cocoa butter, beeswax and polyethylene glycols.

The pharmaceutical compositions of this invention may also be administered topically, especially when the target of treatment includes areas or organs readily accessible by topical application, including diseases of the eye, the skin, or the lower intestinal tract. Suitable topical formulations are readily prepared for each of these areas or organs.

Topical application for the lower intestinal tract can be effected in a rectal suppository formulation

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(see above) or in a suitable enema formulation.
Topically-transdermal patches may also be used.

For topical applications, the pharmaceutical compositions may be formulated in a suitable ointment containing the active component suspended or dissolved in one or more carriers. Carriers for topical administration of the compounds of this invention include, but are not limited to, mineral oil, liquid petrolatum, white petrolatum, propylene glycol, polyoxyethylene, polyoxypropylene compound, emulsifying wax and water. Alternatively, the pharmaceutical compositions can be formulated in a suitable lotion or cream containing the active components suspended or dissolved in one or more pharmaceutically acceptable carriers. Suitable carriers include, but are not limited to, mineral oil, sorbitan monostearate, polysorbate 60, cetyl esters wax, cetearyl alcohol, 2-octyldodecanol, benzyl alcohol and water.

For ophthalmic use, the pharmaceutical compositions may be formulated as micronized suspensions in isotonic, pH adjusted sterile saline, or, preferably, as solutions in isotonic, pH adjusted sterile saline, either with or without a preservative such as benzylalkonium chloride. Alternatively, for ophthalmic uses, the pharmaceutical compositions may be formulated in an ointment such as petrolatum.

The pharmaceutical compositions of this invention may also be administered by nasal aerosol or inhalation. Such compositions are prepared according to techniques well-known in the art of pharmaceutical formulation and may be prepared as solutions in saline, employing benzyl alcohol or other suitable preservatives,

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absorption promoters to enhance bioavailability, fluorocarbons, and/or other conventional solubilizing or dispersing agents.

The amount of p38 inhibitor that may be combined with the carrier materials to produce a single dosage form will vary depending upon the host treated, the particular mode of administration. Preferably, the compositions should be formulated so that a dosage of between 0.01 - 100 mg/kg body weight/day of the inhibitor can be administered to a patient receiving these compositions.

It should also be understood that a specific dosage and treatment regimen for any particular patient will depend upon a variety of factors, including the activity of the specific compound employed, the age, body weight, general health, sex, diet, time of administration, rate of excretion, drug combination, and the judgment of the treating physician and the severity of the particular disease being treated. The amount of inhibitor will also depend upon the particular compound in the composition.

According to another embodiment, the invention provides methods for treating or preventing a p38-mediated condition comprising the step of administering to a patient one of the above-described pharmaceutical compositions. The term "patient", as used herein, means an animal, preferably a human.

Preferably, that method is used to treat or prevent a condition selected from inflammatory diseases, autoimmune diseases, destructive bone disorders, proliferative disorders, infectious diseases, degenerative diseases, allergies, reperfusion/ischemia in

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stroke, heart attacks, angiogenic disorders, organ hypoxia, vascular hyperplasia, cardiac hypertrophy, and thrombin-induced platelet aggregation.

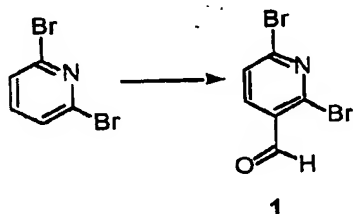
According to another embodiment, the inhibitors
5 of this invention are used to treat or prevent an IL-1, IL-6, IL-8 or TNF-mediated disease or condition. Such conditions are described above.

Depending upon the particular p38-mediated
condition to be treated or prevented, additional drugs,
10 which are normally administered to treat or prevent that condition, may be administered together with the inhibitors of this invention. For example, chemotherapeutic agents or other anti-proliferative agents may be combined with the p38 inhibitors of this
15 invention to treat proliferative diseases.

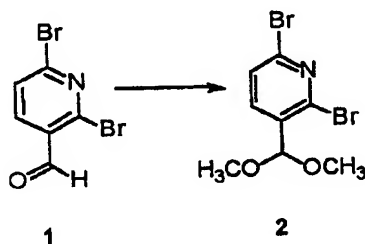
Those additional agents may be administered separately, as part of a multiple dosage regimen, from the p38 inhibitor-containing composition. Alternatively, those agents may be part of a single dosage form, mixed
20 together with the p38 inhibitor in a single composition.

In order that the invention described herein may be more fully understood, the following examples are set forth. It should be understood that these examples are for illustrative purposes only and are not to be
25 construed as limiting this invention in any manner.

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EXAMPLE 1Synthesis of p38 Inhibitor Compound 6

To a solution of LDA (60mmol, 40mLs) at -78°C ,
was added dropwise a solution of 2,6-Dibromopyridine
5 (40mmol, 9.48gms) in THF (30mLs, dried). The mixture was
stirred at -78°C for 20 minutes. Ethyl formate (400mmol,
32.3mLs) was added and stirring was continued at -78°C
for 2 hours. Saturated ammonium chloride (200mLs) was
added and the mixture was warmed to room temperature.
10 The reaction mixture was diluted with ethyl acetate and
the organic layer was washed with aqueous acid and base.
The organic layer was dried and evaporated *in vacuo*. The
resulting material was purified by flash chromatography
on silica gel followed by eluting with 10% ethyl acetate
15 in n-hexane to afford 1 (32mmol, 8.41gms) as a white
solid.

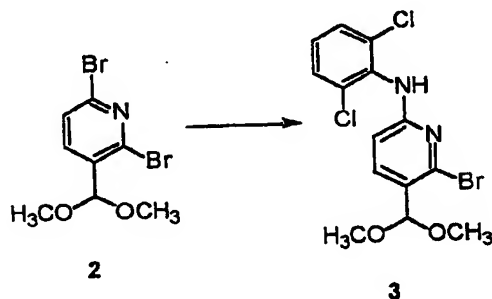


A solution of 1 (13.08mmol, 3.1gms) and
concentrated sulfuric acid (1mL) in methanol (50mL) was
20 refluxed overnight. The reaction mixture was cooled,

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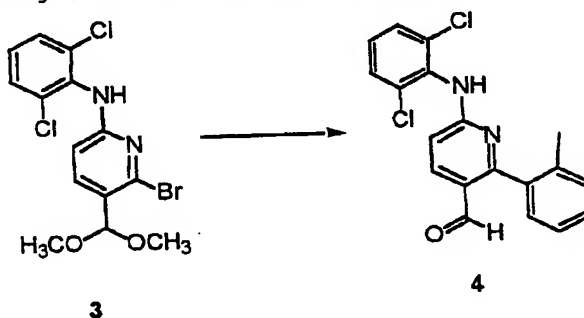
neutralized with aqueous base and extracted into ethyl acetate. Drying and evaporation of the organic layer afforded 2 (11.77mmol, 3.63gms) as an oil.



5 To a solution of t-Butoxide (2.2mmol, 2mLs) was added dropwise a solution of 2,6-Dichloroaniline (1.0mmol, 162mgs) in THF (2mL, dried). The mixture was stirred at room temperature for 20 minutes. A solution of 2 (1.0mmol, 309mgs) in THF (5mLs) was added and stirring

10 was continued for 3 hours. The reaction mixture was diluted with ethyl acetate and the organic layer was washed with aqueous acid and base. The organic layer was dried and evaporated in vacuo. The resulting material was purified by flash chromatography on silica gel

15 followed by eluting with 5% acetone in n-hexane to afford 3 (0.33mmol, 128mgs) as an orange solid.

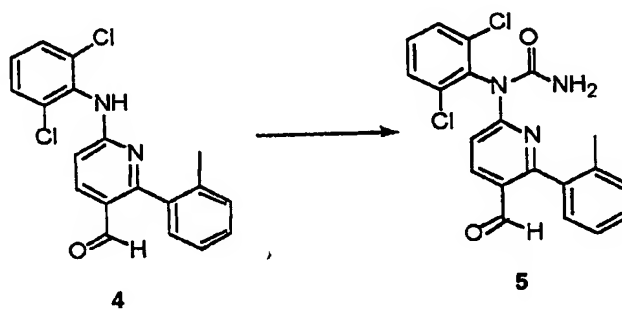


o-Tolylboronic acid (0.34mmol, 46mgs), and 3 (0.20mmol, 80mgs) were dissolved in a toluene/ethanol

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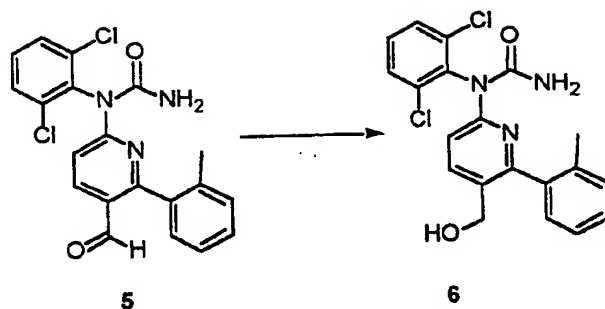
(5/1) mixture. Thallium carbonate (0.5, 235mgs) and tetrakis(triphenylphosphine)palladium (0) (10mgs) was added to the solution and the slurry was allowed to reflux for 30 minutes. The reaction mixture was diluted with ethyl acetate and the organic layer was washed with aqueous acid and base. The organic layer was dried and evaporated in *vacuo*. The resulting material was purified by flash chromatography on silica gel followed by eluting with 5% methanol in methylene chloride to afford 4 (0.17mmol, 61mgs) as a white solid.



A solution of 4 (0.17mmol, 61mgs) and chlorosulfonyl isocyanate (1mmol, 141.5mgs) in methylene chloride (5mLs) was stirred at room temperature overnight. The reaction mixture was diluted with ethyl acetate and the organic layer was washed with aqueous acid and base. The organic layer was dried and evaporated in *vacuo*. The resulting material was purified by flash chromatography on silica gel followed by eluting with 5% acetone in n-hexane to afford 5 (0.12mmol, 46mgs) as a white solid.

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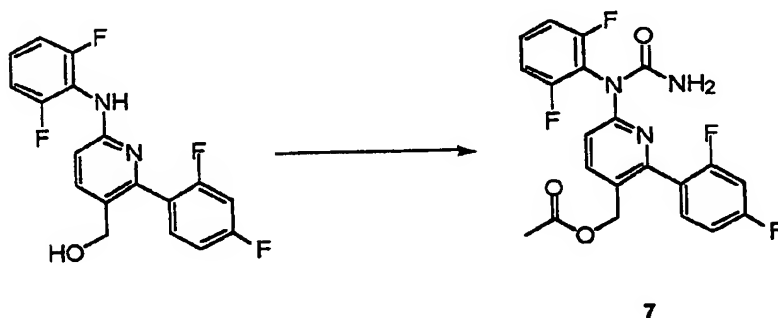


Sodium borohydride (1.0mmol, 39.8 mgs) was added to a solution of 5 (0.12mmol, 46mgs) in methanol (10mLs) and the solution was stirred for 15 minutes. The reaction was quenched with water. The reaction mixture was then diluted with ethyl acetate and the organic layer was washed with aqueous acid and base. The organic layer was dried and evaporated in vacuo. The resulting material was purified by flash chromatography to afford 6 (0.08mmol, 36mgs) as a white solid.

The spectral data for compound 6 was:

^1H NMR (500 MHz, CDCl_3) δ 7.90 (d, 1H), 7.60 (d, 2H), 7.5-7.3 (m, 5H), 6.30 (d, 2H), 4.5 (s, 2H), 2.3 (s, 2H).

15 Synthesis of p38 Inhibitor Compound 7



The amino-alcohol (500 mg, 1.43 mmol), which was prepared in the same manner as 4, was dissolved in

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dichloromethane. Triethylamine (433 mg, 4.29 mmol) was added, followed by acetyl chloride (168 mg, 2.15 mmol). The mixture was stirred at room temperature for one hour, poured into water, and extracted with dichloromethane.

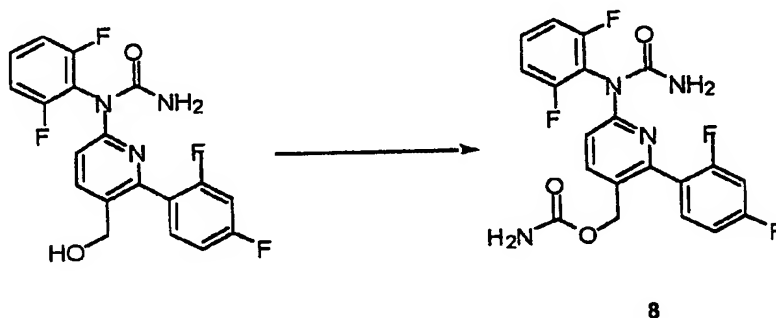
- 5 The organic extract was evaporated *in vacuo* and the residue was dissolved in 10.0 mL of toluene. A 20% solution of phosgene in toluene (5.0 mL) was added and the solution was refluxed for two hours. The solution was cooled and 5.0 mL of concentrated ammonium hydroxide
10 was added, precipitating a white solid. The mixture was poured into water and extracted with toluene. The organic extract was dried (MgSO₄) and evaporated *in vacuo* to afford 205 mg of the urea-acetate 7 as a white solid.

The spectral data for compound 7 was:

- 15 ¹H NMR (500 MHz, CDCl₃) δ 7.80 (d, 1H), 7.62-7.50 (m, 2H), 7.25-7.0 (m, 5H), 6.59 (d, 1H), 5.1 (s, 2H), 2.12 (s, 3H). HRMS showed MH⁺ 434.2 as the major peak.

Synthesis of p38 Inhibitor Compound 8

20



- The urea-alcohol (548 mg, 1.4 mmol), which was prepared in the same manner as 6, was dissolved in 5.0 mL
25 of toluene. A 20% solution of phosgene in toluene (5.0 mL) was added and the solution was refluxed for two

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hours. The solution was cooled and 5.0 mL of concentrated ammonium hydroxide was added, precipitating a white solid. The mixture was poured into water and extracted with toluene. The organic extract was dried (MgSO₄) and evaporated in vacuo to afford 284 mg of the carbamate **8** as a white solid.

The spectral data of compound **8** was:

¹H NMR (500 MHz, CDCl₃) δ 7.77 (d, 1H), 7.55-7.45 (m, 2H), 7.15-6.95 (m, 5H), 6.50 (d, 1H), 5.40 (br s, 2H), 5.00 (s, 2H). HRMS showed MH⁺ 435.1 as the major peak.

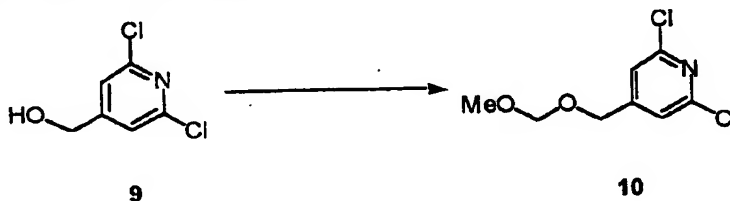
EXAMPLE 2

Synthesis of p38 Inhibitor Compound 16



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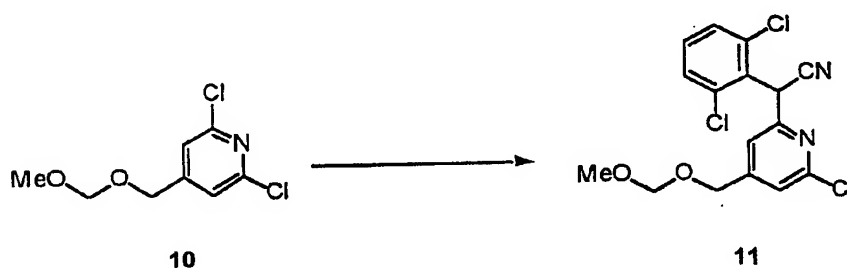
One equivalent of 2,6-dichloropyridine-4-carboxylic acid was dissolved in THF. The solution was cooled to 0°C and one equivalent of borane dimethyl sulfide complex was added. The solution was stirred at room temperature for twelve hours. The mixture was poured into water and extracted with diethyl ether. The ether extract was dried, and evaporated in vacuo to afford **9** in 93% yield.



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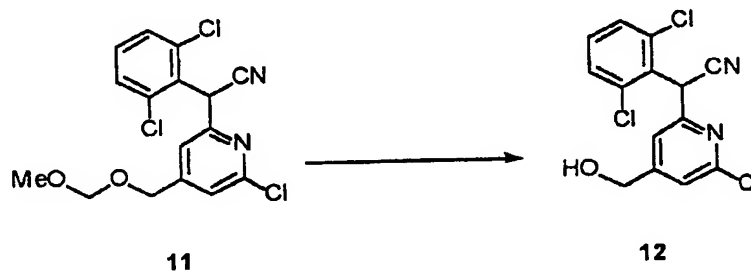
One equivalent of 9 was dissolved in methylene chloride. One equivalent of methyl chloromethyl ether was added, followed by the addition of one equivalent of ethyl diisopropylamine. The reaction was stirred at room temperature for several hours, poured into water and extracted with a water-immiscible solvent. The extract was dried and evaporated in vacuo to afford 10 in 86% yield.



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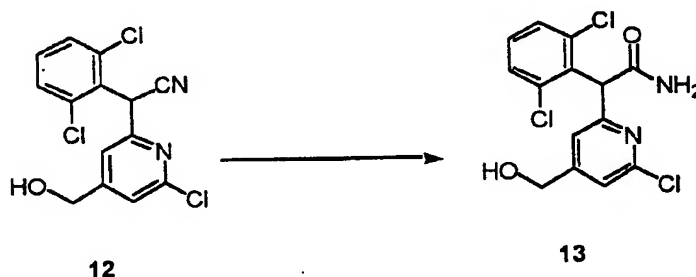
One equivalent of potassium t-butoxide was added to a solution of one equivalent of 2,6-dichlorophenyl acetonitrile in THF at room temperature. The mixture was stirred at room temperature for thirty minutes, and a solution of the dichloropyridine 10 in THF was added. After stirring for 1.5 hours, the mixture was poured into aqueous ammonium chloride and extracted with ethyl acetate. The extract was dried and evaporated in vacuo. The residue was purified by flash chromatography to afford 11 in 79% yield as a white powder.

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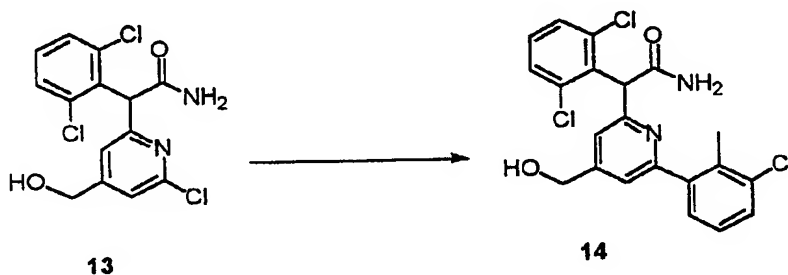
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The acetal 11 was mixed with concentrated hydrochloric acid and stirred for several hours. The mixture was extracted with a water-immiscible organic solvent. The extract was washed with saturated aqueous NaHCO_3 , dried, and evaporated in vacuo to afford 12.



The nitrile 12 was mixed with concentrated sulfuric acid and heated to 100°C for several minutes. The mixture was cooled, poured onto ice, and filtered to afford 13.

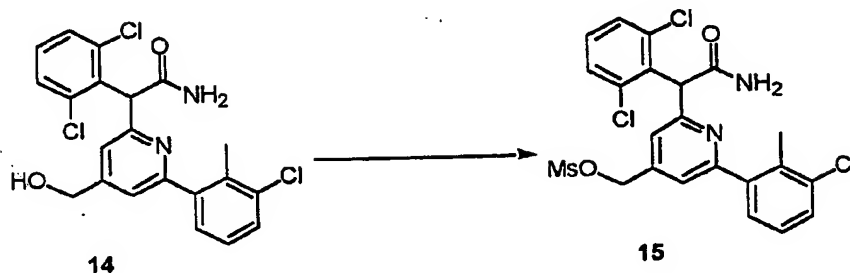


One equivalent of the chloropyridine 13 was dissolved in 1,2-dimethoxyethane. One equivalent of 3-chloro-2-methylphenylboronic acid was added. A solution of one equivalent of sodium carbonate in water was added along with a catalytic amount of tetrakis (triphenylphosphine) palladium (0). The mixture was heated to 80°C for several hours. The mixture was poured into water and extracted with a water-immiscible organic

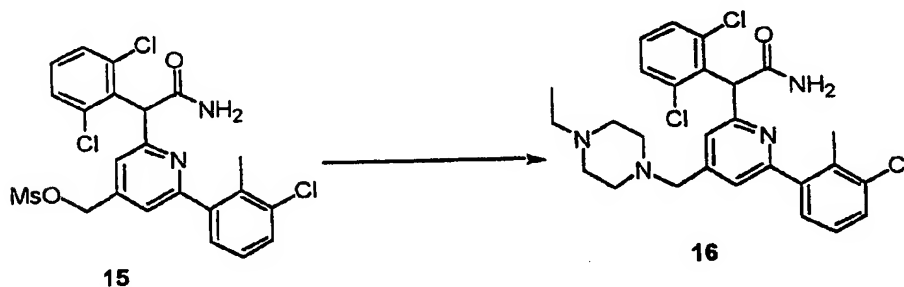
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solvent. The extract was dried, evaporated in vacuo and purified by flash chromatography to afford **14**.



5 One equivalent of the alcohol **14** was dissolved in THF. The solution was cooled to 0°C and one equivalent of methanesulfonyl chloride was added following by one equivalent of triethylamine. The solution was stirred for several hours, poured into
10 water, and extracted with a water-immiscible solvent. The extract was dried and evaporated in vacuo to afford the crude mesylate **15**.



15 One equivalent of the methanesulfonyl ester **15** was dissolved in THF. The solution was cooled to 0°C and one equivalent of N-ethyl piperazine was added following by one equivalent of triethylamine. The solution was stirred for several hours, poured into water, and
20 extracted with a water-immiscible solvent. The extract was dried, evaporated, and purified by flash chromatography to afford the pure amine **16**.

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The spectral data for compound 16 is:

¹H NMR (500 MHz, CDCl₃) δ 9.85 (br s, 1H), 7.47 (dd, 1H),
7.42 (d, 1H), 7.27 (m, 5H), 6.75 (s, 1H), 5.95 (s, 1H),
5.7 (br s, 1H), 3.5 (ABq, 2H), 2.5-2.3 (m, 10H), 2.3 (s,
5 3H), 1.2 (t, 3H).

EXAMPLE 2

Cloning of p38 Kinase in Insect Cells

Two splice variants of human p38 kinase, CSBP1 and CSBP2, have been identified. Specific oligonucleotide primers were used to amplify the coding
10 region of CSBP2 cDNA using a HeLa cell library (Stratagene) as a template. The polymerase chain reaction product was cloned into the pET-15b vector (Novagen). The baculovirus transfer vector, pVL-(His)6-p38 was constructed by subcloning a XbaI-BamHI fragment
15 of pET15b-(His)6-p38 into the complementary sites in plasmid pVL1392 (Pharmingen).

The plasmid pVL-(His)6-p38 directed the synthesis of a recombinant protein consisting of a 23-residue peptide (MGSSHHHHHSSGLVPRGSHMLE, where LVPRGS
20 represents a thrombin cleavage site) fused in frame to the N-terminus of p38, as confirmed by DNA sequencing and by N-terminal sequencing of the expressed protein. Monolayer culture of *Spodoptera frugiperda* (Sf9) insect cells (ATCC) was maintained in TNM-FH medium (Gibco BRL)
25 supplemented with 10% fetal bovine serum in a T-flask at 27°C. Sf9 cells in log phase were co-transfected with linear viral DNA of Autographa californica nuclear polyhedrosis virus (Pharmingen) and transfer vector pVL-(His)6-p38 using Lipofectin (Invitrogen). The individual

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recombinant baculovirus clones were purified by plaque assay using 1% low melting agarose.

EXAMPLE 3

Expression and Purification of Recombinant p38 Kinase

Trichoplusia ni (Tn-368) High-Five™ cells (Invitrogen) were grown in suspension in Excel-405 protein-free medium (JRH Bioscience) in a shaker flask at 27°C. Cells at a density of 1.5×10^6 cells/ml were infected with the recombinant baculovirus described above at a multiplicity of infection of 5. The expression level of recombinant p38 was monitored by immunoblotting using a rabbit anti-p38 antibody (Santa Cruz Biotechnology). The cell mass was harvested 72 hours after infection when the expression level of p38 reached its maximum.

Frozen cell paste from cells expressing the (His)₆-tagged p38 was thawed in 5 volumes of Buffer A (50 mM NaH₂PO₄, pH 8.0, 200 mM NaCl, 2mM β-Mercaptoethanol, 10% Glycerol and 0.2 mM PMSF). After mechanical disruption of the cells in a microfluidizer, the lysate was centrifuged at 30,000 x g for 30 minutes. The supernatant was incubated batchwise for 3-5 hours at 4°C with Talon™ (Clontech) metal affinity resin at a ratio of 1 ml of resin per 2-4 mgs of expected p38. The resin was settled by centrifugation at 500 x g for 5 minutes and gently washed batchwise with Buffer A. The resin was slurried and poured into a column (approx. 2.6 x 5.0 cm) and washed with Buffer A + 5 mM imidazole.

The (His)₆-p38 was eluted with Buffer A + 100 mM imidazole and subsequently dialyzed overnight at 4°C against 2 liters of Buffer B, (50 mM HEPES, pH 7.5, 25 mM

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β -glycerophosphate, 5% glycerol, 2mM DTT). The His₆ tag was removed by addition of at 1.5 units thrombin (Calbiochem) per mg of p38 and incubation at 20°C for 2-3 hours. The thrombin was quenched by addition of 0.2 mM PMSF and then the entire sample was loaded onto a 2 ml benzamidine agarose (American International Chemical) column.

The flow through fraction was directly loaded onto a 2.6 x 5.0 cm Q-Sepharose (Pharmacia) column previously equilibrated in Buffer B + 0.2 mM PMSF. The p38 was eluted with a 20 column volume linear gradient to 0.6M NaCl in Buffer B. The eluted protein peak was pooled and dialyzed overnight at 4°C vs. Buffer C (50 mM HEPES pH 7.5, 5% glycerol, 50 mM NaCl, 2 mM DTT, 0.2 mM PMSF).

The dialyzed protein was concentrated in a Centriprep (Amicon) to 3-4 ml and applied to a 2.6 x 100 cm Sephacryl S-100HR (Pharmacia) column. The protein was eluted at a flow rate of 35 ml/hr. The main peak was pooled, adjusted to 20 mM DTT, concentrated to 10-80 mgs/ml and frozen in aliquots at -70°C or used immediately.

EXAMPLE 4

Activation of p38

p38 was activated by combining 0.5 mg/ml p38 with 0.005 mg/ml DD-double mutant MKK6 in Buffer B + 10mM MgCl₂, 2mM ATP, 0.2mM Na₂VO₄ for 30 minutes at 20°C. The activation mixture was then loaded onto a 1.0 x 10 cm MonoQ column (Pharmacia) and eluted with a linear 20 column volume gradient to 1.0 M NaCl in Buffer B. The

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activated p38 eluted after the ADP and ATP. The activated p38 peak was pooled and dialyzed against buffer B + 0.2mM Na₂VO₄ to remove the NaCl. The dialyzed protein was adjusted to 1.1M potassium phosphate by addition of a 4.0M stock solution and loaded onto a 1.0 x 10 cm HIC (Rainin Hydropore) column previously equilibrated in Buffer D (10% glycerol, 20mM β -glycerophosphate, 2.0mM DTT) + 1.1MK₂HPO₄. The protein was eluted with a 20 column volume linear gradient to Buffer D + 50mM K₂HPO₄. The double phosphorylated p38 eluted as the main peak and was pooled for dialysis against Buffer B + 0.2mM Na₂VO₄. The activated p38 was stored at -70°C.

EXAMPLE 5

p38 Inhibition Assays

A. Inhibition of Phosphorylation of EGF Receptor Peptide

This assay was carried out in the presence of 10 mM MgCl₂, 25 mM β -glycerophosphate, 10% glycerol and 100 mM HEPES buffer at pH 7.6. For a typical IC₅₀ determination, a stock solution was prepared containing all of the above components and activated p38 (5 nM). The stock solution was aliquotted into vials. A fixed volume of DMSO or inhibitor in DMSO (final concentration of DMSO in reaction was 5%) was introduced to each vial, mixed and incubated for 15 minutes at room temperature. EGF receptor peptide, KRELVEPLTPSGEAPNQALLR, a phosphoryl acceptor in p38-catalyzed kinase reaction (1), was added to each vial to a final concentration of 200 μ M. The kinase reaction was initiated with ATP (100 μ M) and the vials were incubated at 30°C. After 30 minutes, the

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reactions were quenched with equal volume of 10% trifluoroacetic acid (TFA).

The phosphorylated peptide was quantified by HPLC analysis. Separation of phosphorylated peptide from the unphosphorylated peptide was achieved on a reverse phase column (Deltapak, 5 μ m, C18 100D, Part no. 011795) with a binary gradient of water and acetonitrile, each containing 0.1% TFA. IC_{50} (concentration of inhibitor yielding 50% inhibition) was determined by plotting the percent (%) activity remaining against inhibitor concentration.

B. Inhibition of ATPase Activity

This assay is carried out in the presence of 10 mM $MgCl_2$, 25 mM β -glycerophosphate, 10% glycerol and 100 mM HEPES buffer at pH 7.6. For a typical K_i determination, the K_m for ATP in the ATPase activity of activated p38 reaction is determined in the absence of inhibitor and in the presence of two concentrations of inhibitor. A stock solution is prepared containing all of the above components and activated p38 (60 nM). The stock solution is aliquotted into vials. A fixed volume of DMSO or inhibitor in DMSO (final concentration of DMSO in reaction was 2.5%) is introduced to each vial, mixed and incubated for 15 minutes at room temperature. The reaction is initiated by adding various concentrations of ATP and then incubated at 30°C. After 30 minutes, the reactions are quenched with 50 μ l of EDTA (0.1 M, final concentration), pH 8.0. The product of p38 ATPase activity, ADP, is quantified by HPLC analysis.

Separation of ADP from ATP is achieved on a reversed phase column (Supelcosil, LC-18, 3 μ m, part no.

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5-8985) using a binary solvent gradient of following composition: Solvent A - 0.1 M phosphate buffer containing 8 mM tetrabutylammonium hydrogen sulfate (Sigma Chemical Co., catalogue no. T-7158), Solvent B -
5 Solvent A with 30% methanol.

Ki is determined from the rate data as a function of inhibitor and ATP concentrations.

p38 inhibitors of this invention will inhibit the ATPase activity of p38.

10

C. Inhibition of IL-1, TNF, IL-6 and IL-8
Production in LPS-Stimulated PBMCs

Inhibitors were serially diluted in DMSO from a 20 mM stock. At least 6 serial dilutions were prepared.
15 Then 4x inhibitor stocks were prepared by adding 4 μ l of an inhibitor dilution to 1 ml of RPMI1640 medium/10% fetal bovine serum. The 4x inhibitor stocks contained inhibitor at concentrations of 80 μ M, 32 μ M, 12.8 μ M, 5.12 μ M, 2.048 μ M, 0.819 μ M, 0.328 μ M, 0.131 μ M, 0.052
20 μ M, 0.021 μ M etc. The 4x inhibitor stocks were pre-warmed at 37°C until use.

Fresh human blood buffy cells were separated from other cells in a Vacutainer CPT from Becton & Dickinson (containing 4 ml blood and enough DPBS without
25 Mg^{2+}/Ca^{2+} to fill the tube) by centrifugation at 1500 x g for 15 min. Peripheral blood mononuclear cells (PBMCs), located on top of the gradient in the Vacutainer, were removed and washed twice with RPMI1640 medium/10% fetal bovine serum. PBMCs were collected by centrifugation at
30 500 x g for 10 min. The total cell number was determined using a Neubauer Cell Chamber and the cells were adjusted to a concentration of 4.8×10^6 cells/ml in cell culture

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medium (RPMI1640 supplemented with 10% fetal bovine serum).

Alternatively, whole blood containing an anti-coagulant was used directly in the assay.

5 100 μ l of cell suspension or whole blood were placed in each well of a 96-well cell culture plate. Then 50 μ l of the 4x inhibitor stock was added to the cells. Finally, 50 μ l of a lipopolysaccharide (LPS) working stock solution (16 ng/ml in cell culture medium) was added to give a final concentration of 4 ng/ml LPS in the assay. The total assay volume of the vehicle control was also adjusted to 200 μ l by adding 50 μ l cell culture medium. The PBMC cells or whole blood were then incubated overnight (for 12-15 hours) at 37° C/5% CO₂ in a humidified atmosphere.

10

15

The next day the cells were mixed on a shaker for 3-5 minutes before centrifugation at 500 x g for 5 minutes. Cell culture supernatants were harvested and analyzed by ELISA for levels of IL-1b (R & D Systems, Quantikine kits, #DBL50), TNF- α (BioSource, #KHC3012), IL-6 (Endogen, #EH2-IL6) and IL-8 (Endogen, #EH2-IL8) according to the instructions of the manufacturer. The ELISA data were used to generate dose-response curves from which IC₅₀ values were derived.

20

25 Results for the kinase assay ("kinase"; subsection A, above), IL-1 and TNF in LPS-stimulated PBMCs ("cell") and IL-1, TNF and IL-6 in whole blood ("WB") for various p38 inhibitors of this invention are shown in Table 7 below:

Table 7.

Compound	M.W.	Kinase IC ₅₀ (uM)	Cell IL-1 IC ₅₀ (uM)	Cell TNF IC ₅₀ (uM)	WB IL-1 IC ₅₀ (uM)	WB TNF IC ₅₀ (uM)	WB IL-6 IC ₅₀ (uM)
17	402.28	0.056	0.021	0.14	0.42	0.064	0.25
18	436.32	0.002	0.02	0.05	0.118	0.055	0.18
19	387.36	0.027	0.027	0.01	0.057	0.09	0.075

Other p38 inhibitors of this invention will also inhibit phosphorylation of EGF receptor peptide, and will inhibit the production of IL-1, TNF and IL-6, as well as IL-8, in LPS-stimulated PBMCs or in whole blood.

D. Inhibition of IL-6 and IL-8
Production in IL-1-Stimulated PBMCs

This assay is carried out on PBMCs exactly the same as above except that 50 µl of an IL-1b working stock solution (2 ng/ml in cell culture medium) is added to the assay instead of the (LPS) working stock solution.

Cell culture supernatants are harvested as described above and analyzed by ELISA for levels of IL-6 (Endogen, #EH2-IL6) and IL-8 (Endogen, #EH2-IL8) according to the instructions of the manufacturer. The ELISA data are used to generate dose-response curves from which IC₅₀ values were derived.

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E. Inhibition of LPS-Induced
Prostaglandin Endoperoxide Synthase-2
(PGHS-2, or COX-2) Induction in PBMCs

Human peripheral mononuclear cells (PBMCs) are
5 isolated from fresh human blood buffy coats by
centrifugation in a Vacutainer CPT (Becton & Dickinson).
15 $\times 10^6$ cells are seeded in a 6-well tissue culture dish
containing RPMI 1640 supplemented with 10% fetal bovine
serum, 50U/ml penicillin, 50 μ g/ml streptomycin, and 2 mM
10 L-glutamine. Compounds are added at 0.2, 2.0 and 20 μ M
final concentrations in DMSO. LPS is then added at a
final concentration of 4 ng/ml to induce enzyme
expression. The final culture volume is 10 ml/well.

After overnight incubation at 37°C, 5% CO₂, the
15 cells are harvested by scraping and subsequent
centrifugation, the supernatant is removed, and the cells
are washed twice in ice-cold DPBS (Dulbecco's phosphate
buffered saline, BioWhittaker). The cells are lysed on
ice for 10 min in 50 μ l cold lysis buffer (20 mM Tris-
20 HCl, pH 7.2, 150 mM NaCl, 1% Triton-X-100, 1% deoxycholic
acid, 0.1% SDS, 1 mM EDTA, 2% aprotinin (Sigma), 10 μ g/ml
pepstatin, 10 μ g/ml leupeptin, 2 mM PMSF, 1 mM
benzamidine, 1 mM DTT) containing 1 μ l Benzonase (DNase
from Merck). The protein concentration of each sample is
25 determined using the BCA assay (Pierce) and bovine serum
albumin as a standard. Then the protein concentration of
each sample is adjusted to 1 mg/ml with cold lysis
buffer. To 100 μ l lysate an equal volume of 2xSDS PAGE
loading buffer is added and the sample is boiled for 5
30 min. Proteins (30 μ g/lane) are size-fractionated on 4-
20% SDS PAGE gradient gels (Novex) and subsequently
transferred onto nitrocellulose membrane by

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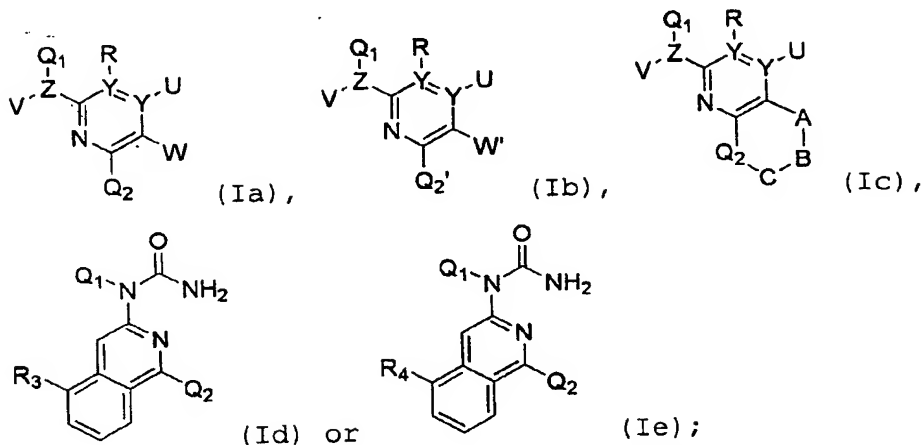
electrophoretic means for 2 hours at 100 mA in Towbin transfer buffer (25 mM Tris, 192 mM glycine) containing 20% methanol. After transfer, the membrane is pretreated for 1 hour at room temperature with blocking buffer (5% non-fat dry milk in DPBS supplemented with 0.1% Tween-20) and washed 3 times in DPBS/0.1% Tween-20. The membrane is incubated overnight at 4°C with a 1: 250 dilution of monoclonal anti-COX-2 antibody (Transduction Laboratories) in blocking buffer. After 3 washes in DPBS/0.1% Tween-20, the membrane is incubated with a 1:1000 dilution of horseradish peroxidase-conjugated sheep antiserum to mouse Ig (Amersham) in blocking buffer for 1 h at room temperature. Then the membrane is washed again 3 times in DPBS/0.1% Tween-20. An ECL detection system (SuperSignal™ CL-HRP Substrate System, Pierce) is used to determine the levels of expression of COX-2.

While we have hereinbefore presented a number of embodiments of this invention, it is apparent that our basic construction can be altered to provide other embodiments which utilize the methods of this invention.

CLAIMS

We claim:

1. A compound of the formula:



wherein each of Q_1 and Q_2 are independently selected from a phenyl or 5-6 membered aromatic heterocyclic ring system, or a 8-10 membered bicyclic ring system comprising aromatic carbocyclic rings, aromatic heterocyclic rings or a combination of an aromatic carbocyclic ring and an aromatic heterocyclic ring;

the rings that make up Q_1 are substituted with 1 to 4 substituents, each of which is independently selected from halo; C_1 - C_3 alkyl optionally substituted with NR'_2 , OR' , CO_2R' or $CONR'_2$; O -(C_1 - C_3)-alkyl optionally substituted with NR'_2 , OR' , CO_2R' or $CONR'_2$; NR'_2 ; OCF_3 ; CF_3 ; NO_2 ; CO_2R' ; $CONR'$; SR' ; $S(O_2)N(R')_2$; SCF_3 ; CN ; $N(R')C(O)R^4$; $N(R')C(O)OR^4$; $N(R')C(O)C(O)R^4$; $N(R')S(O_2)R^4$; $N(R')R^4$; $N(R^4)_2$; OR^4 ; $OC(O)R^4$; $OP(O)_3H_2$; or $N=C-N(R')_2$;

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the rings that make up Q_2 are optionally substituted with up to 4 substituents, each of which is independently selected from halo; C_1 - C_3 straight or branched alkyl optionally substituted with NR'_2 , OR' , CO_2R' , $S(O_2)N(R')_2$, $N=C-N(R')_2$, R^3 , or $CONR'_2$; $O-(C_1-C_3)$ -alkyl; $O-(C_1-C_3)$ -alkyl optionally substituted with NR'_2 , OR' , CO_2R' , $S(O_2)N(R')_2$, $N=C-N(R')_2$, R^3 , or $CONR'_2$; NR'_2 ; OCF_3 ; CF_3 ; NO_2 ; CO_2R' ; $CONR'$; R^3 ; OR^3 ; NR^3 ; SR^3 ; $C(O)R^3$; $C(O)N(R')R^3$; $C(O)OR^3$; SR' ; $S(O_2)N(R')_2$; SCF_3 ; $N=C-N(R')_2$; or CN ;

Q_2' is selected from phenyl or a 5-6 member aromatic heterocyclic ring optionally substituted with 1-3 substituents, each of which is independently selected from halogen; C_1 - C_3 alkyl optionally substituted with NR'_2 , OR' , CO_2R' , $CONR'_2$, or $O-P(O_3)H_2$; $O-(C_2-C_3)$ -alkyl optionally substituted with NR'_2 , OR' , CO_2R' , $CONR'_2$, or $OP(O_3)H_2$; OCF_3 ; CF_3 ; OR^4 ; $O-CO_2R^4$; $O-P(O_3)H_2$; CO_2R' ; $CONR'$; SR' ; $S(O_2)N(R')_2$; SCF_3 ; CN ; $N(R')C(O)R^4$; $N(R')C(O)OR^4$; $N(R')C(O)C(O)R^4$; $N(R')S(O_2)R^4$; $N(R')R^4$; $N(R^4)_2$; OR^4 ; $OC(O)R^4$; $OP(O)_3H_2$; or $N=C-N(R')_2$; provided that Q_2' is not phenyl optionally substituted 1 to 3 substituents independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl;

R' is selected from hydrogen; (C_1-C_3) -alkyl; (C_2-C_3) -alkenyl or alkynyl; phenyl or phenyl substituted with 1 to 3 substituents independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl; or a 5-6 membered heterocyclic ring system optionally substituted with 1 to 3 substituents

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independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl;

R^3 is selected from 5-8 membered aromatic or non-aromatic carbocyclic or heterocyclic ring systems each optionally substituted with R' , R^4 , $-C(O)R'$, $-C(O)R^4$, $-C(O)OR^4$ or $-J$; or an 8-10 membered bicyclic ring system comprising aromatic carbocyclic rings, aromatic heterocyclic rings or a combination of an aromatic carbocyclic ring and an aromatic heterocyclic ring each optionally substituted with R' , R^4 , $-C(O)R'$, $-C(O)R^4$, $-C(O)OR^4$ or $-J$;

R^4 is (C_1-C_4) -straight or branched alkyl optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, or $SO_2N(R^2)_2$; or a 5-6 membered carbocyclic or heterocyclic ring system optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, or $SO_2N(R^2)_2$;

R^5 is selected from hydrogen; (C_1-C_3) -alkyl optionally substituted with R^3 ; (C_2-C_3) -alkenyl or alkynyl each optionally substituted with R^3 ; phenyl or phenyl substituted with 1 to 3 substituents independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl; or a 5-6 membered heterocyclic ring system optionally substituted with 1 to 3 substituents independently selected from halo, methoxy, cyano, nitro, amino, hydroxy, methyl or ethyl;

W is selected from $N(R^2)SO_2-N(R^2)_2$; $N(R^2)SO_2-N(R^2)(R^3)$; $N(R^2)C(O)-OR^2$; $N(R^2)C(O)-N(R^2)_2$; $N(R^2)C(O)-N(R^2)(R^3)$; $N(R^2)C(O)-R^2$; $N(R^2)_2$; $C(O)-R^2$; $CH(OH)-R^2$; $C(O)-N(R^2)_2$; $C(O)-OR^2$; J ; or (C_1-C_4) straight or branched alkyl optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, R^3 , $SO_2N(R^2)_2$, $OC(O)R^2$, $OC(O)R'$, $OC(O)N(R^2)_2$, $-N(R^4)(R^5)$, $-$

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$C(O)N(R^5)(R^2)$, $-C(O)R^5$, $-N(R^2)C(O)N(R^2)(R^5)$, $-NC(O)OR^5$, $-OC(O)N(R^2)(R^5)$, or $-J$; a 5-6 membered carbocyclic or heterocyclic ring system optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, or $SO_2N(R^2)_2$; or a 8-10 membered carbocyclic or heterocyclic ring system optionally substituted with $N(R')_2$, OR' , CO_2R' , $CON(R')_2$, or $SO_2N(R^2)_2$; provided that W is not an R^3 substituted C_1 alkyl;

W' is selected from $N(R^2)-SO_2-Q_2$; $N(R^2)-CO_2-Q_2$; $N(R^2)-C(O)-Q_2$; $N(R^2)(Q_2)$; $C(O)-Q_2$; CO_2-Q_2 ; $C(O)N(R^2)(Q_2)$; $C(R^2)_2Q_2$;

each R is independently selected from hydrogen, $-R^2$, $-N(R^2)_2$, $-OR^2$, SR^2 , $-C(O)-N(R^2)_2$, $-S(O_2)-N(R^2)_2$, $-C(O)-OR^2$ or $-C(O)R^2$ wherein two adjacent R are optionally bound to one another and, together with each Y to which they are respectively bound, form a 4-8 membered carbocyclic or heterocyclic ring;

R^2 is selected from hydrogen, (C_1-C_3) -alkyl, or (C_1-C_3) -alkenyl; each optionally substituted with $-N(R')_2$, $-OR'$, SR' , $-C(O)-N(R')_2$, $-S(O_2)-N(R')_2$, $-C(O)-OR'$, $-NSO_2R^4$, $-NSO_2R^3$, $-C(O)N(R')(R^3)$, $-NC(O)R^4$, $-N(R')(R^3)$, $-N(R')(R^4)$, $-C(O)R^3$, $-C(O)N(R')(R^4)$, $-N(R^4)_2$, $-C(O)N=C(NH)_2$ or R^3 ;

Y is N or C;

Z is CH, N, $C(OCH_3)$, $C(CH_3)$, $C(NH_2)$, $C(OH)$ or $C(F)$;

U is selected from R or W;

V is selected from $-C(O)NH_2$, $-P(O)(NH_2)_2$, or $-SO_2NH_2$;

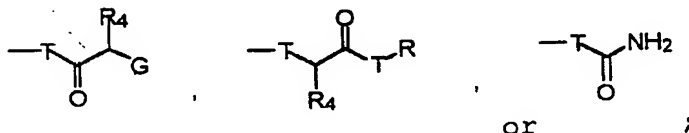
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A, B, and C are independently selected from -O-, -CHR'-, -CHR⁴-, -NR'-, -NR⁴- or -S-;

J is a (C₁-C₄) straight chain or branched alkyl derivative substituted with 1-3 substituents selected from D, -T-C(O)R', or -OPO₃H₂;

D is selected from the group



T is either O or NH; and

G is either NH₂ or OH.

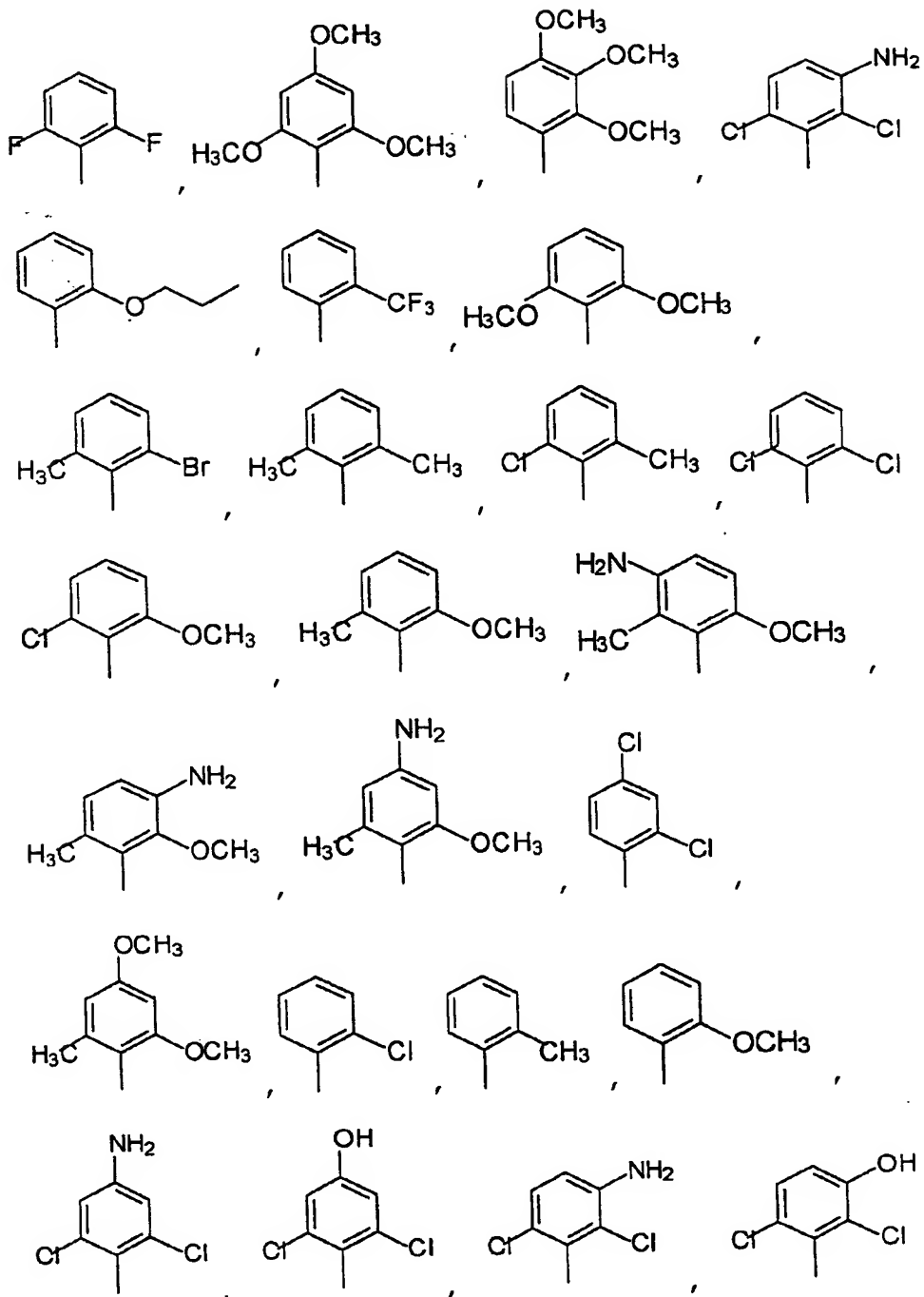
2. The compound according to claim 1, wherein Q₁ is selected from phenyl or pyridyl containing 1 to 3 substituents independently selected from chloro, fluoro, bromo, -CH₃, -OCH₃, -OH, -CF₃, -OCF₃, -O(CH₂)₂CH₃, NH₂, 3,4-methylenedioxy, -N(CH₃)₂, -NH-S(O)₂-phenyl, -NH-C(O)O-CH₂-4-pyridine, -NH-C(O)CH₂-morpholine, -NH-C(O)CH₂-N(CH₃)₂, -NH-C(O)CH₂-piperazine, -NH-C(O)CH₂-pyrrolidine, -NH-C(O)C(O)-morpholine, -NH-C(O)C(O)-piperazine, -NH-C(O)C(O)-pyrrolidine, -O-C(O)CH₂-N(CH₃)₂, or -O-(CH₂)₂-N(CH₃)₂ and wherein at least one of said substituents is in the ortho position.

3. The compound according to claim 2, wherein Q₁ contains at least two substituents, both of which are in the ortho position.

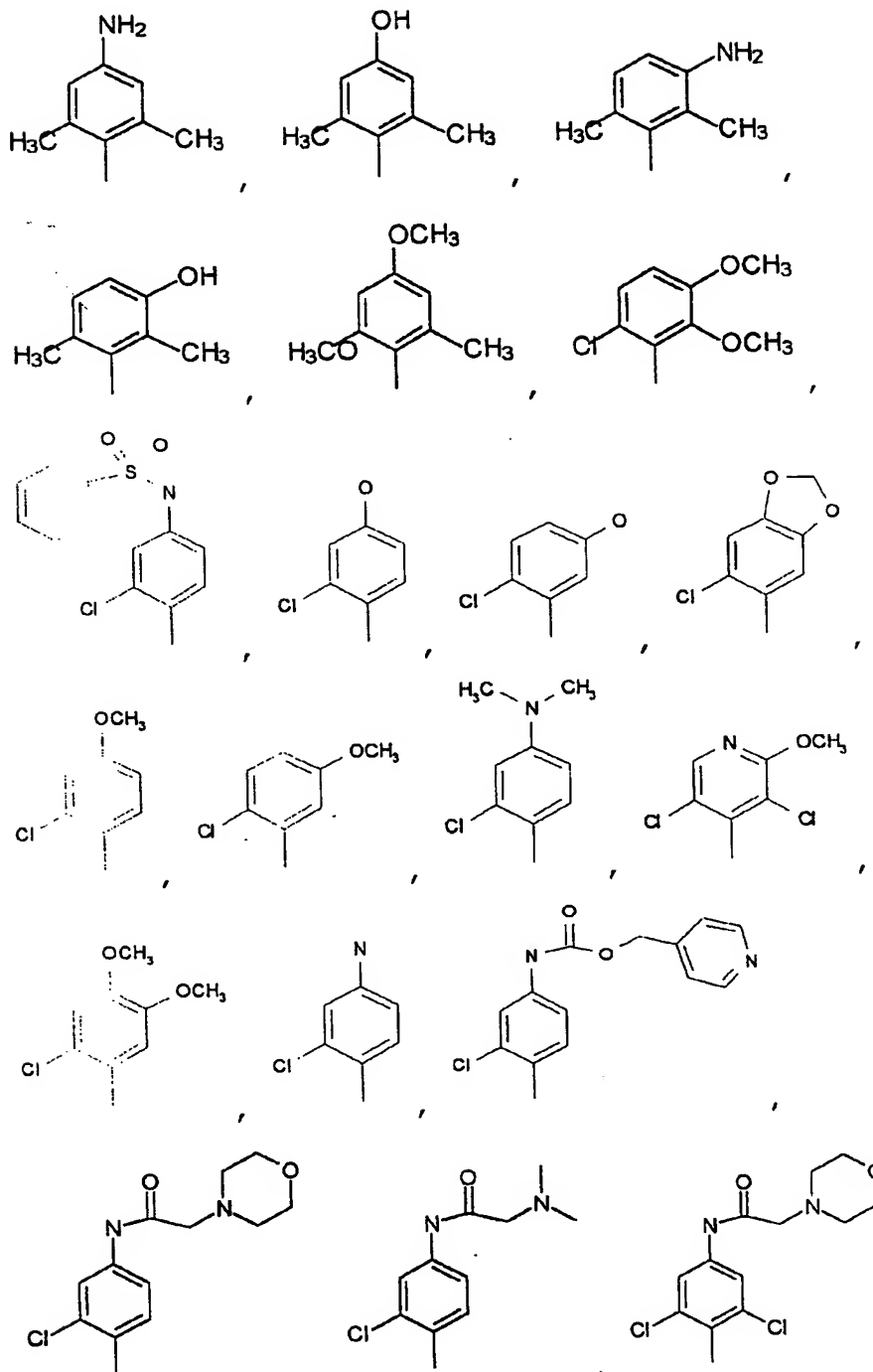
4. The compound according to claim 2, wherein Q₁ is selected from:

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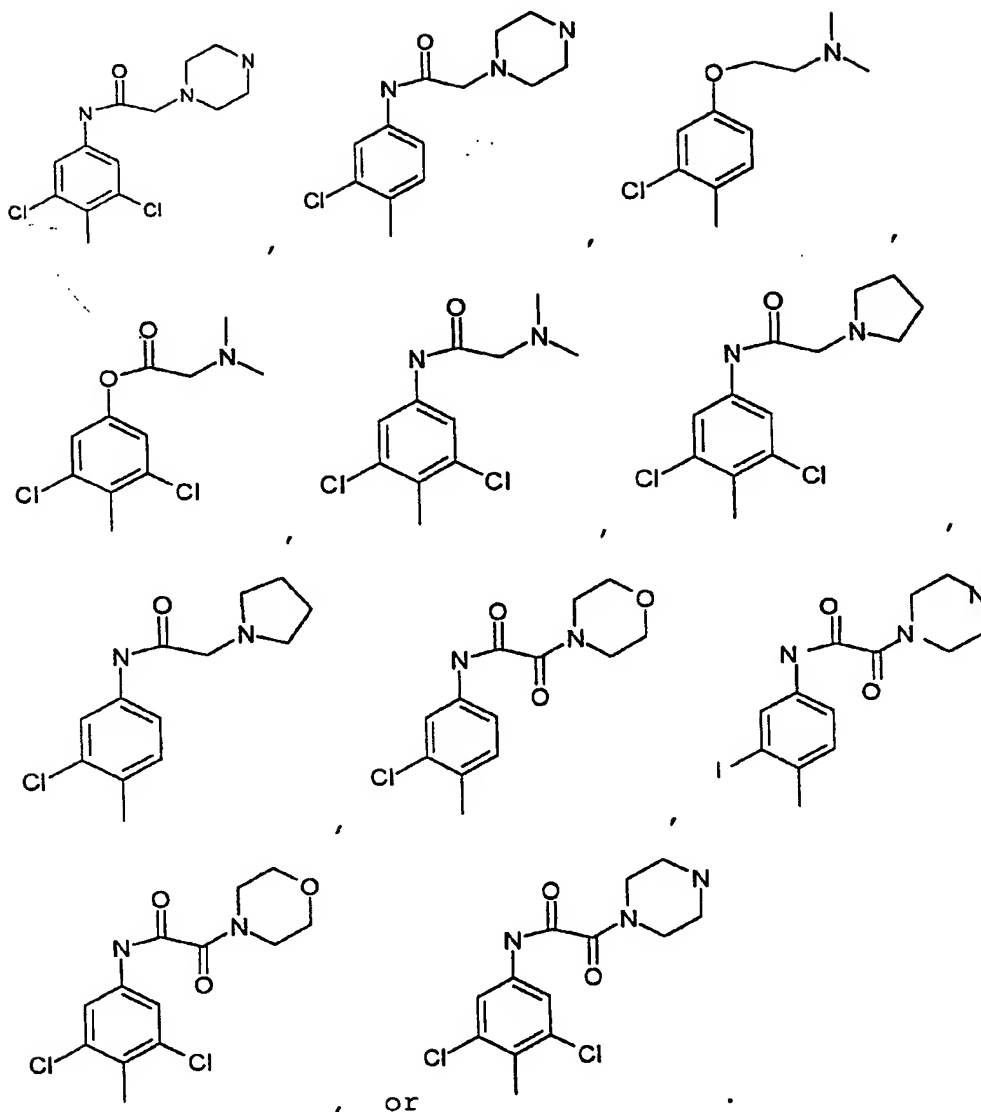
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5. The compound according to claim 4, wherein Q_1 is selected from 2-fluoro-6-trifluoromethylphenyl, 2,6-difluorophenyl, 2,6-dichlorophenyl, 2-chloro-4-hydroxyphenyl, 2-chloro-4-aminophenyl, 2,6-dichloro-4-aminophenyl, 2,6-dichloro-3-aminophenyl, 2,6-dimethyl-4-aminophenyl, 2-methoxy-3,5-dichloro-4-pyridyl, 2-

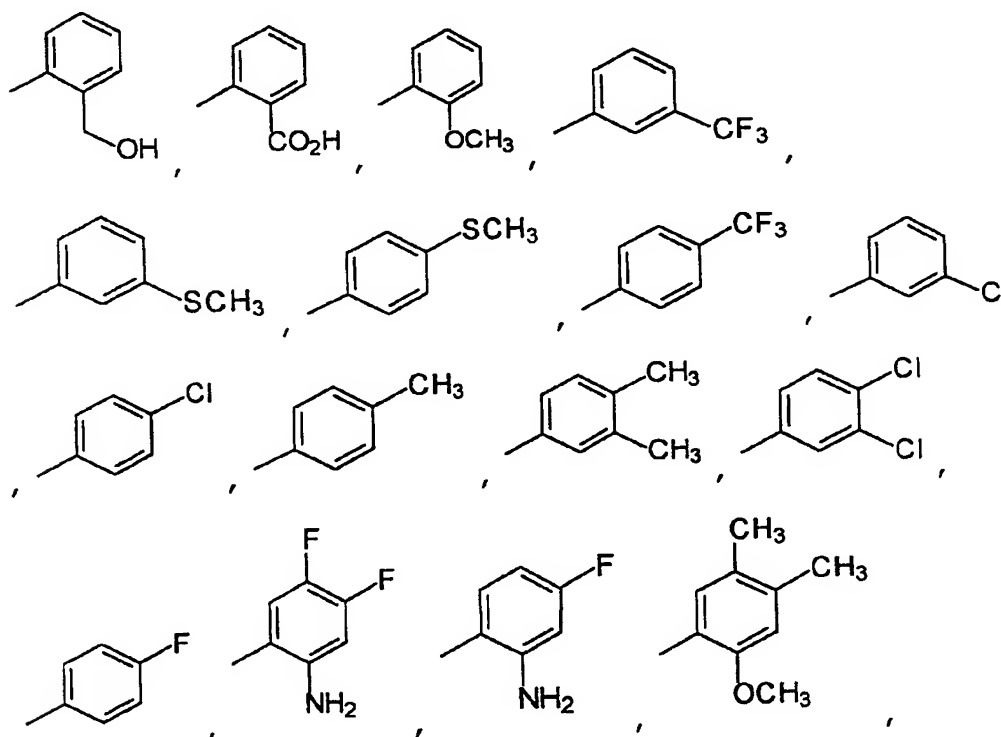
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chloro-4,5 methylenedioxy phenyl, or 2-chloro-4-(N-2-morpholino-acetamido)phenyl.

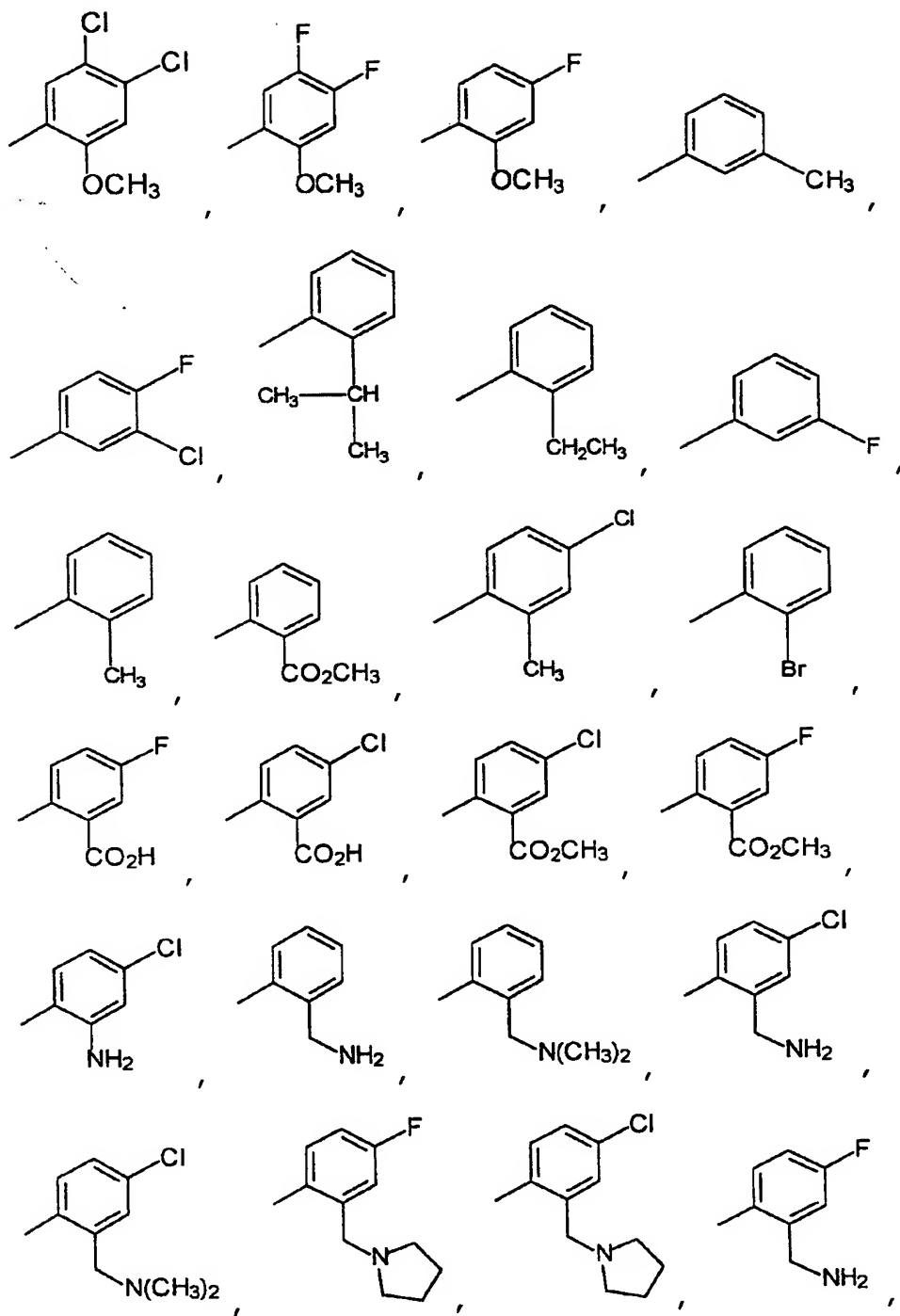
6. The compound according to claim 1, wherein Q_2 is selected from phenyl, pyridyl or naphthyl and wherein Q_2 optionally contains up to 3 substituents, each of which is independently selected from chloro, fluoro, bromo, methyl, ethyl, isopropyl, $-OCH_3$, $-OH$, $-NH_2$, $-CF_3$, $-OCF_3$, $-SCH_3$, $-OCH_3$, $-C(O)OH$, $-C(O)OCH_3$, $-CH_2NH_2$, $-N(CH_3)_2$, $-CH_2$ -pyrrolidine and $-CH_2OH$.

7. The compound according to claim 6, wherein Q_2 is selected from:

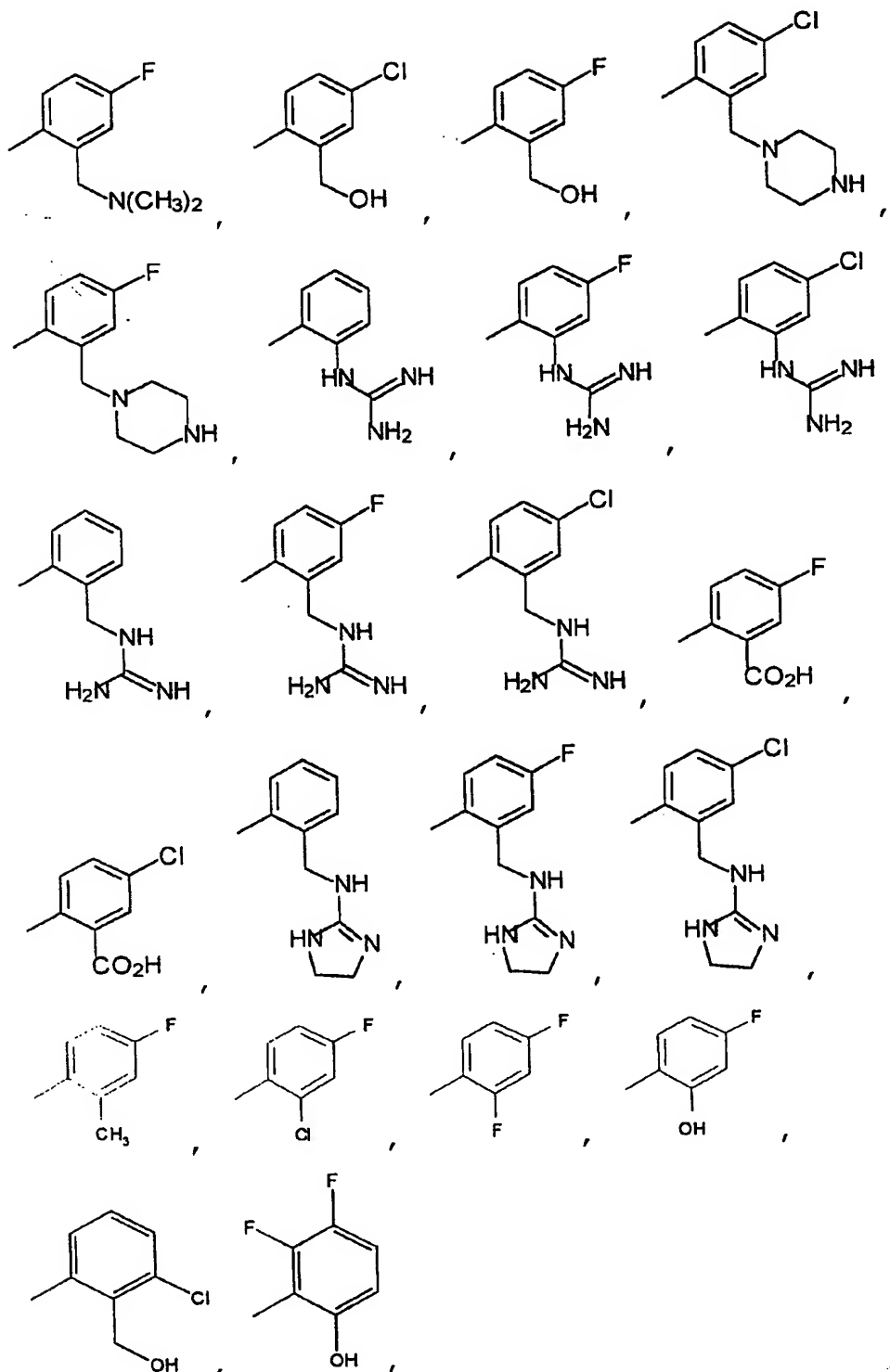


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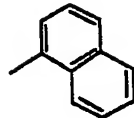
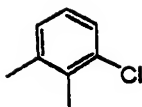
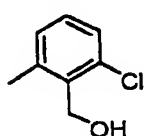
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, unsubstituted 2-pyridyl or unsubstituted phenyl.

8. The compound according to claim 7, wherein Q₂ is selected from phenyl, 2-isopropylphenyl, 3,4-dimethylphenyl, 2-ethylphenyl, 3-fluorophenyl, 2-methylphenyl, 3-chloro-4-fluorophenyl, 3-chlorophenyl, 2-carbomethoxyphenyl, 2-carboxyphenyl, 2-methyl-4-chlorophenyl, 2-bromophenyl, 2-pyridyl, 2-methylenedihydroxyphenyl, 4-fluorophenyl, 2-methyl-4-fluorophenyl, 2-chloro-4-fluorophenyl, 2,4-difluorophenyl, 2-hydroxy-4-fluorophenyl or 2-methylenedihydroxy-4-fluorophenyl, 1-naphthyl, 3-chloro-2-methylenedihydroxy, 3-chloro-2-methyl, or 4-fluoro-2-methyl.

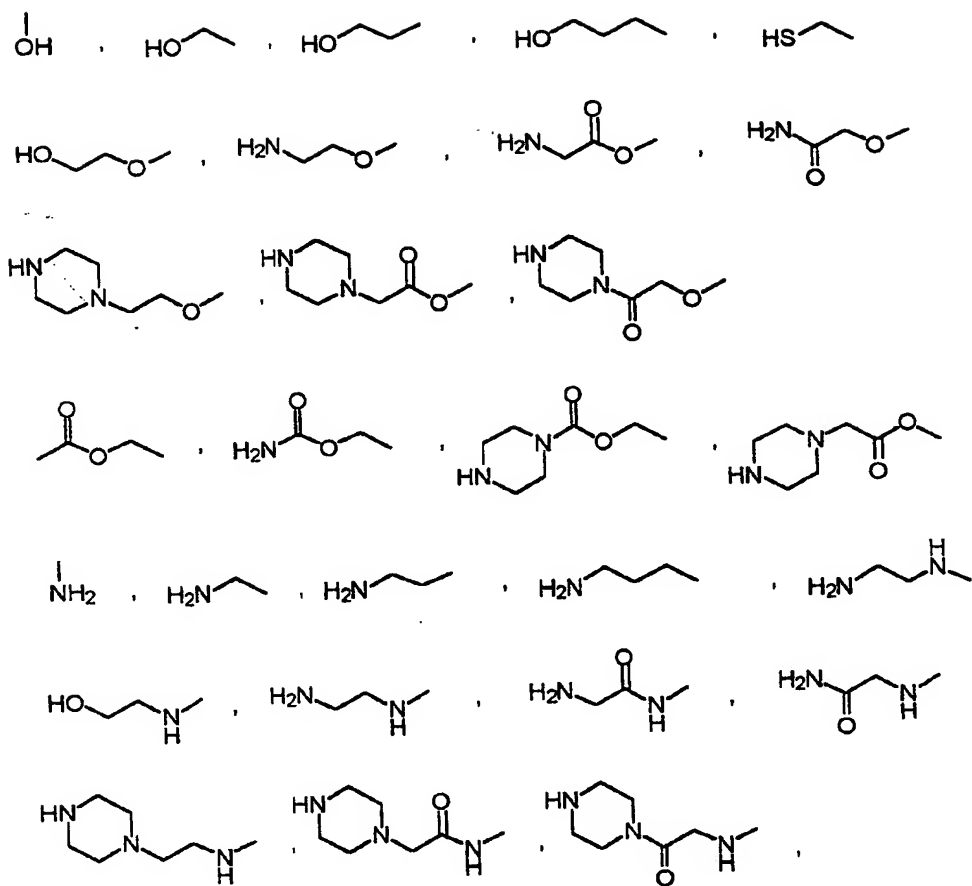
9. The compound according to claim 1, wherein each Y is C.

10. The compound according to claim 9, wherein each R and U attached to Y is independently selected from hydrogen or methyl.

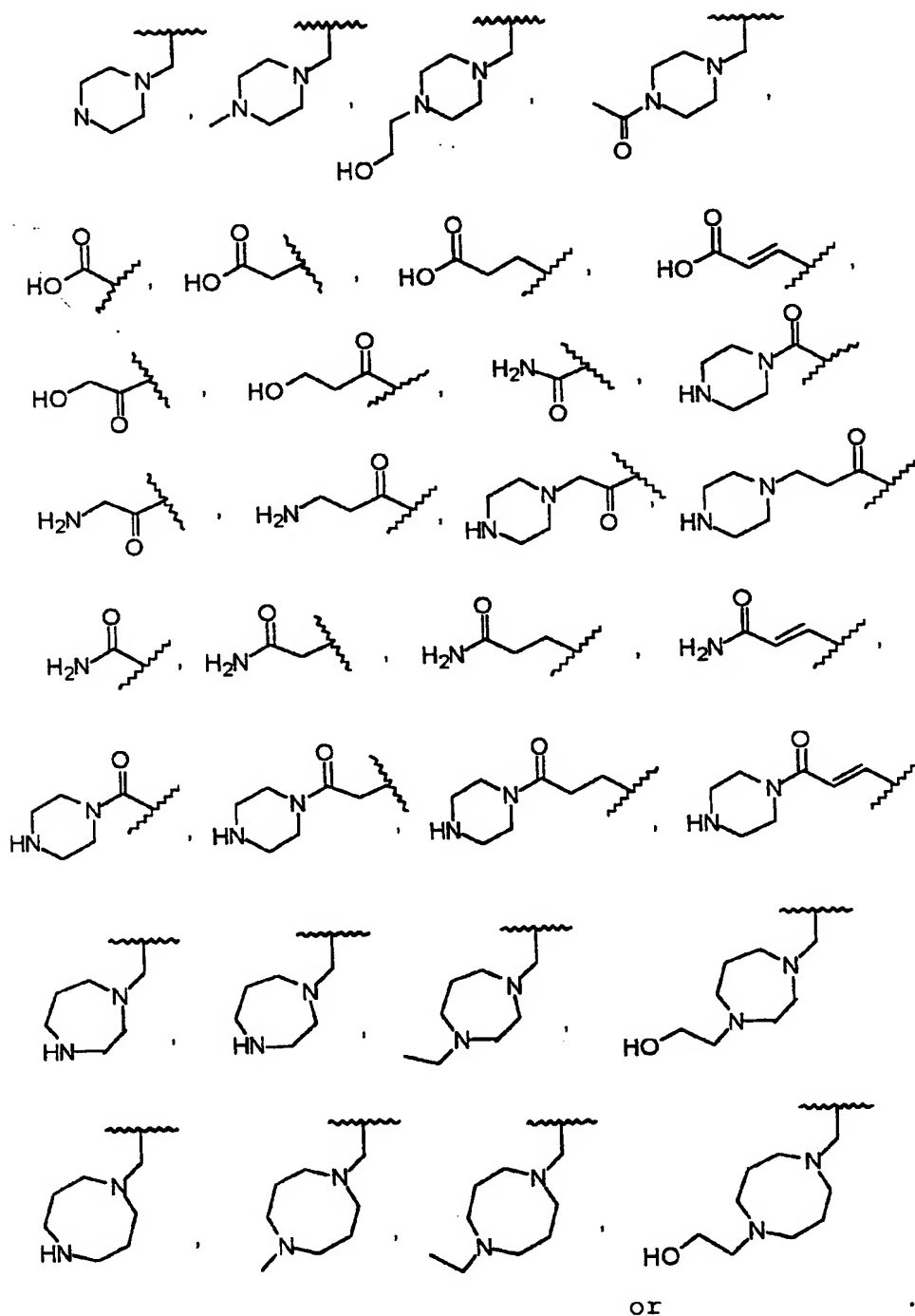
11. The compound according to claim 1, wherein U, W or both U and W are a 0-4 atom chain terminating in an alcohol, amine, carboxylic acid, ester, amide or heterocycle.

12. The compound according to claim 11, wherein U, W or both U and W are selected from:

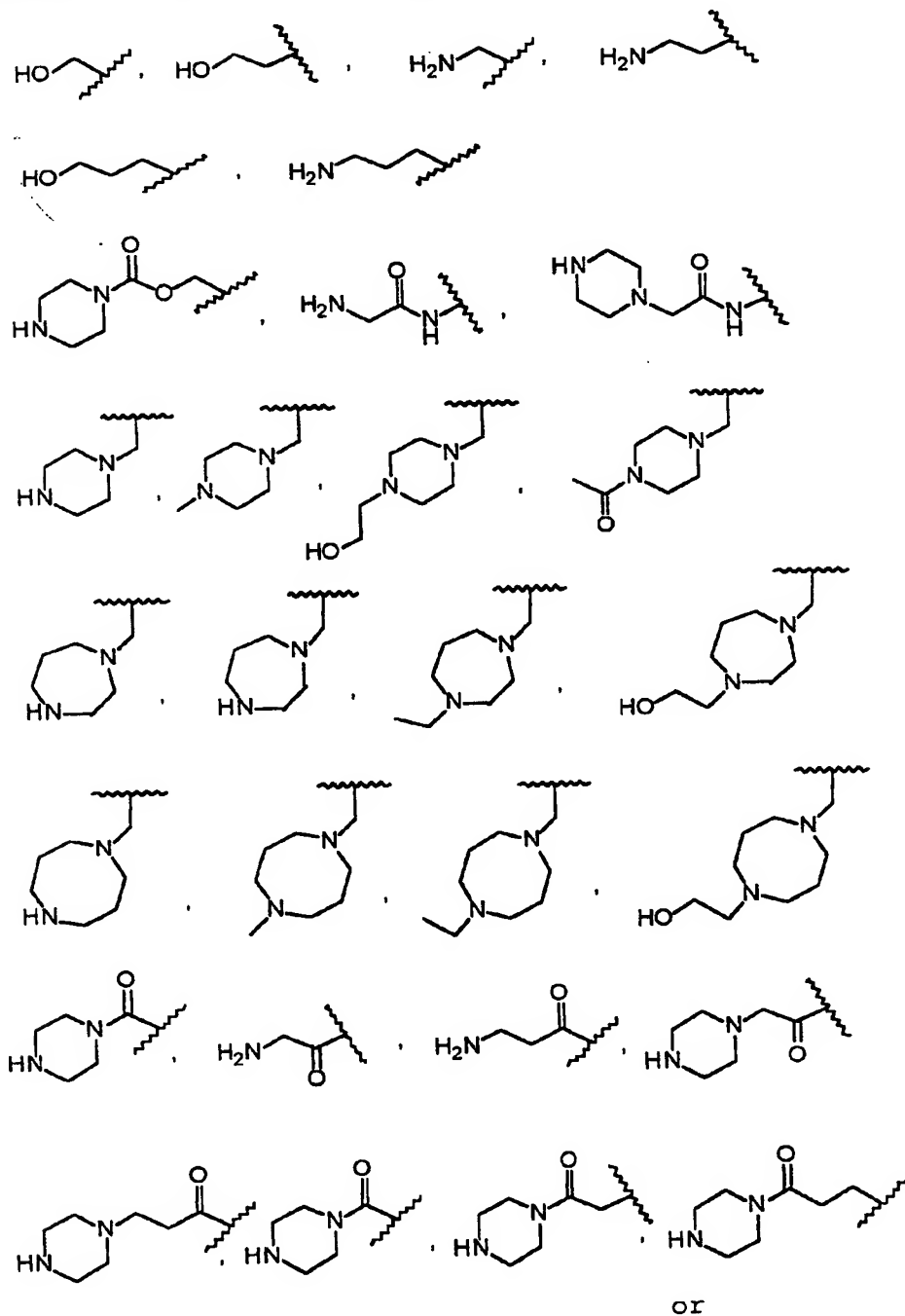
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13. The compound according to claim 12,
wherein U, W, or both U and W are selected from:

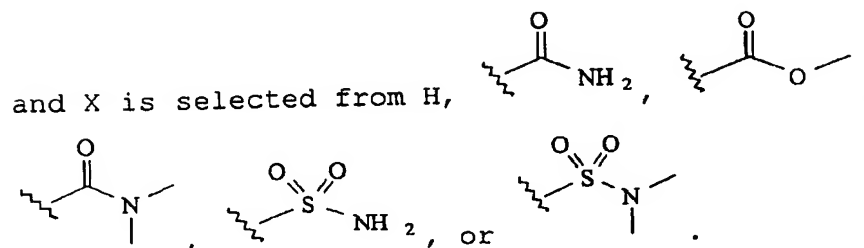
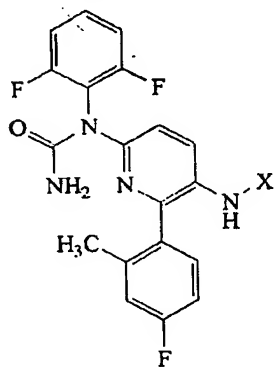


or

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14. The compound according to claim 1, wherein the compound is selected from any one of the compounds depicted in Tables 1 to 6.

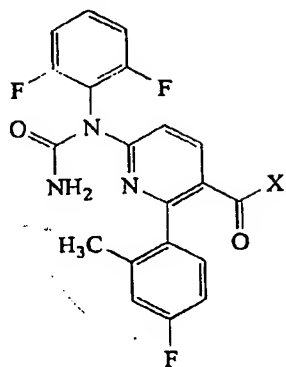
15. The compound according to claim 1, wherein the compound is



16. The compound according to claim 1, wherein the compound is

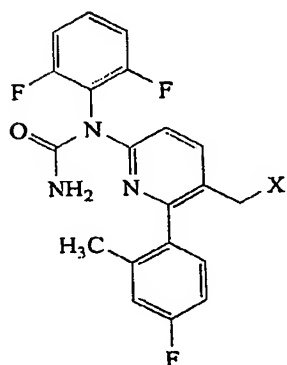
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and X is selected from NH_2 or $\text{N}(\text{CH}_3)_2$;

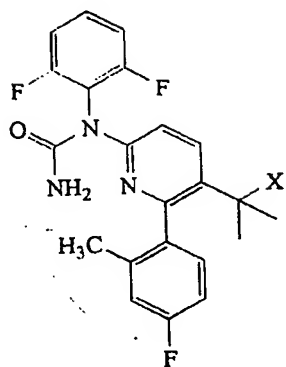
17. The compound according to claim 1, wherein the compound is



and X is selected from OH , NH_2 , or $\text{N}(\text{CH}_3)_2$.

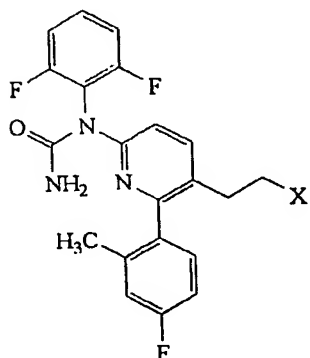
18. The compound according to claim 1, wherein the compound is

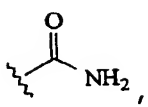
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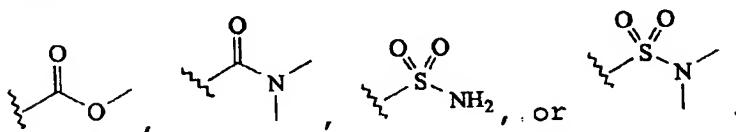


and X is selected from OH, NH₂, or N(CH₃)₂.

19. The compound according to claim 1, wherein the compound is



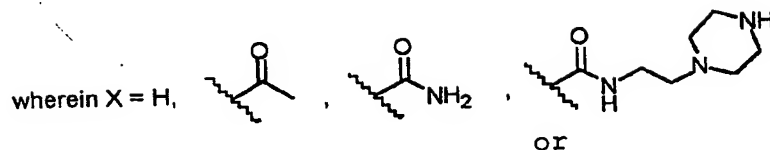
and X is selected from OH, NH₂, N(CH₃)₂, ,



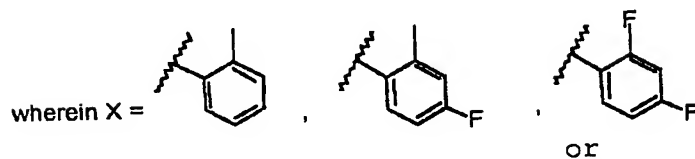
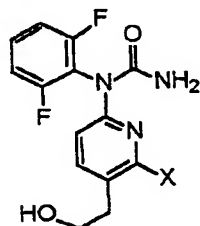
20. The compound according to claim 1, wherein the compound is

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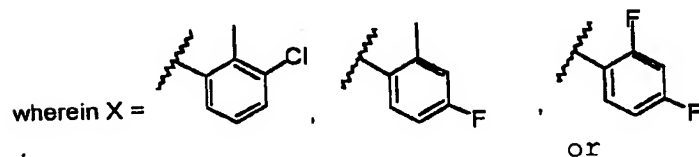
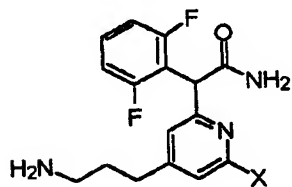
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21. The compound according to claim 1, wherein the compound is



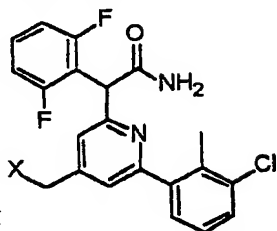
22. The compound according to claim 1, wherein the compound is



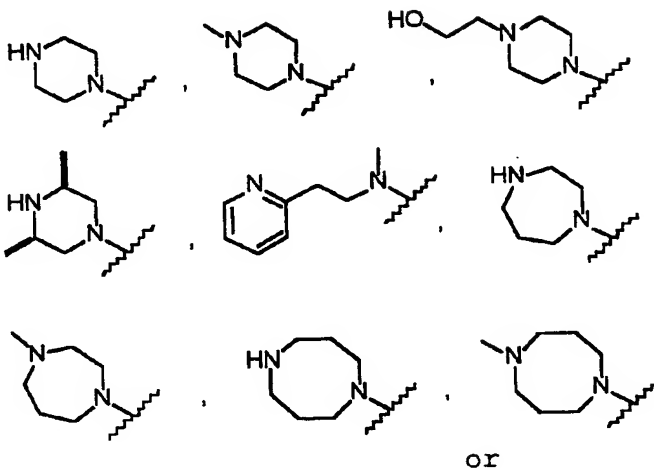
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23. The compound according to claim 1, wherein the compound is

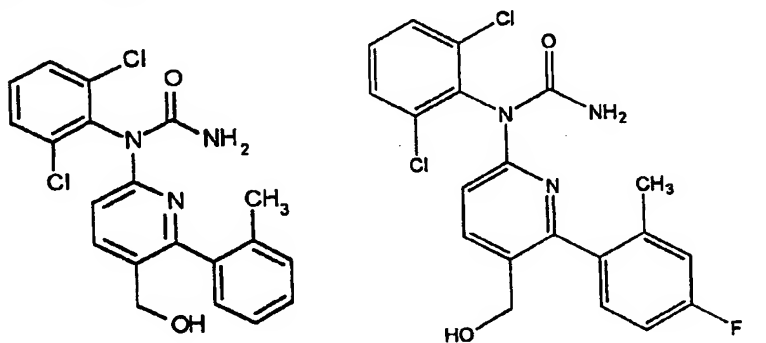


wherein X is



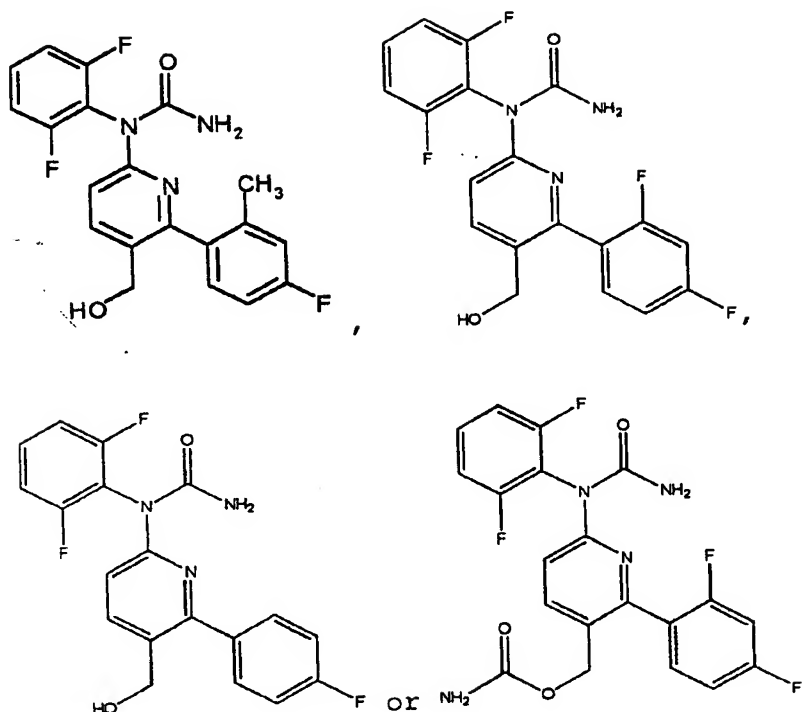
or

24. The compound according to claim 1, wherein said compound is selected from any one of



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25. A pharmaceutical composition comprising an amount of a compound according to any one of claims 1 to 24 effective to inhibit p38, and a pharmaceutically acceptable carrier.

26. A method of treating or preventing inflammatory diseases, autoimmune diseases, destructive bone disorders, proliferative disorders, infectious diseases, neurodegenerative diseases, allergies, reperfusion/ischemia in stroke, heart attacks, angiogenic disorders, organ hypoxia, vascular hyperplasia, cardiac hypertrophy, thrombin-induced platelet aggregation or conditions associated with prostaglandin endoperoxidase synthase-2 in a patient, said method comprising administering to said patient a composition according to claim 25.

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27. The method according to claim 26, wherein said method is used to treat or prevent an inflammatory disease selected from acute pancreatitis, chronic pancreatitis, asthma, allergies, or adult respiratory distress syndrome.

28. The method according to claim 26, wherein said method is used to treat or prevent an autoimmune disease selected from glomerulonephritis, rheumatoid arthritis, systemic lupus erythematosus, scleroderma, chronic thyroiditis, Graves' disease, autoimmune gastritis, diabetes, autoimmune hemolytic anemia, autoimmune neutropenia, thrombocytopenia, atopic dermatitis, chronic active hepatitis, myasthenia gravis, multiple sclerosis, inflammatory bowel disease, ulcerative colitis, Crohn's disease, psoriasis, or graft vs. host disease.

29. The method according to claim 26, wherein said method is used to treat or prevent a destructive bone disorders selected from osteoarthritis, osteoporosis or multiple myeloma-related bone disorder.

30. The method according to claim 26, wherein said method is used to treat or prevent a proliferative disease selected from acute myelogenous leukemia, chronic myelogenous leukemia, metastatic melanoma, Kaposi's sarcoma, or multiple myeloma.

31. The method according to claim 26, wherein said method is used to treat or prevent an infectious

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disease selected from sepsis, septic shock, or Shigellosis.

32. The method according to claim 26, wherein said method is used to treat or prevent a viral disease selected from acute hepatitis infection, HIV infection or CMV retinitis.

33. The method according to claim 26, wherein said method is used to treat or prevent a neurodegenerative disease selected from Alzheimer's disease, Parkinson's disease, cerebral ischemia or neurodegenerative disease caused by traumatic injury.

34. The method according to claim 26, wherein said method is used to treat or prevent ischemia/reperfusion in stroke or myocardial ischemia, renal ischemia, heart attacks, organ hypoxia or thrombin-induced platelet aggregation.

35. The method according to claim 26, wherein said method is used to treat or prevent a condition associated with prostaglandin endoperoxide synthase-2 selected from edema, fever, analgesia or pain.

36. The method according to claim 35, wherein said pain is selected from neuromuscular pain, headache, cancer pain, dental pain or arthritis pain.

37. The method according to claim 26, wherein said method is used to treat or prevent an angiogenic

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disorder selected from solid tumors, ocular
neovascularization, or infantile haemangiomas.

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INTERNATIONAL SEARCH REPORT

International Application No
PCT/US 99/10291

A. CLASSIFICATION OF SUBJECT MATTER

IPC 6 C07D213/56 A61K31/44 C07D213/75 C07D213/74 C07D401/06
C07D213/73

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 6 C07D A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	EP 0 337 944 A (CIBA GEIGY AG) 18 October 1989 (1989-10-18) see claim 1 and examples (e.g. p.53)	1-3, 6, 7
A	WO 97 33883 A (SMITHKLINE BEECHAM CORP ; GALLAGHER TIMOTHY F (US); THOMPSON SUSAN) 18 September 1997 (1997-09-18) claims 1, 27	1-37
A	EP 0 337 943 A (CIBA GEIGY AG) 18 October 1989 (1989-10-18) see claim 1 and examples	1-3, 6, 7
P, X	WO 98 27098 A (GALULLO VINCENT P ; SALITURO FRANCESCO GERALD (US); BEMIS GUY W (US) 25 June 1998 (1998-06-25) page 35; claim 3	1-37

☐ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

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"G" document member of the same patent family

Date of the actual completion of the international search

8 September 1999

Date of mailing of the international search report

17/09/1999

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Steendijk, M

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/US 99/10291

Patent document cited in search report		Publication date	Patent family member(s)	Publication date
EP 0337944	A	18-10-1989	AT 92050 T	15-08-1993
			ES 2058588 T	01-11-1994
			JP 2006477 A	10-01-1990
			US 4973690 A	27-11-1990
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EP 0337943	A	18-10-1989	CA 1314546 A	16-03-1993
			JP 1308265 A	12-12-1989
			US 4966622 A	30-10-1990
			US 5159078 A	27-10-1992
WO 9827098	A	25-06-1998	AU 5610598 A	15-07-1998